

**UNITED STATES DISTRICT COURT
EASTERN DISTRICT OF PENNSYLVANIA**

IN RE: NATIONAL FOOTBALL LEAGUE
PLAYERS' CONCUSSION INJURY
LITIGATION

No. 2:12-md-02323-AB
MDL No. 2323

Kevin Turner and Shawn Wooden,
*on behalf of themselves and
others similarly situated,*

Plaintiffs,

v.

National Football League and
NFL Properties, LLC,
successor-in-interest to
NFL Properties, Inc.,

Defendants.

Civil Action No. 2:14-cv-00029-AB

THIS DOCUMENT RELATES TO:
ALL ACTIONS

SUPPLEMENTAL DECLARATION OF SAM GANDY, M.D., PH.D.

Sam Gandy, M.D., Ph.D., affirms under penalty of perjury the truth of the following facts:

1. I am the Mount Sinai Professor of Alzheimer's Disease Research, Professor of Neurology and Psychiatry, Associate Director of the Mount Sinai Alzheimer's Disease Research Center in New York City, and Chairman Emeritus of the National Medical and Scientific Advisory Council of the Alzheimer's Association.

2. On October 14, 2014, I submitted a declaration in support of the October 6, 2014 Objection of Sean Morey, Alan Faneca, Ben Hamilton, Robert Royal, Roderick Cartwright, Jeff Rohrer, and Sean Considine. *See* Dkt. No. 6232-1. I reaffirm the statements made in that Declaration.

3. I did not receive any financial compensation from any source, including any plaintiffs' lawyer in this case, for preparing this Supplemental Declaration.

4. I understand that on November 19, 2014, the Court held a fairness hearing during which attorneys for the NFL, the plaintiff class, and various objectors presented argument on the fairness, adequacy, and reasonableness of the Settlement.

5. I also understand that during those arguments, Co-Lead Class Counsel Christopher Seeger responded to my October 14 declaration. I have reviewed the transcript of the statements Mr. Seeger made about me and my research during the fairness hearing.

6. At the fairness hearing, Mr. Seeger quoted me as writing that "there have been no prospective studies and clinical and neuropsychological characterization of CTE is yet to be properly developed." *See* Tr. 195:6-9. That quote is taken from an article I authored with others in *Molecular Neurodegeneration* entitled "Chronic Traumatic Encephalopathy: Clinical-Biomarker Correlations and Current Concepts in Pathogenesis." That article is attached as Exhibit A. Nothing in that article undermines or contradicts the statements in my October 14 declaration.

7. Mr. Seeger pulls this quote out of context and ignores what *is known* about CTE.

8. First, that statement notes only the present absence of widely accepted criteria for making a *clinical* diagnosis of CTE. A clinical diagnosis or a clinically accepted diagnosis is a diagnosis of CTE in a living person made by a clinician.

9. A neuropathological diagnosis, by contrast, is made by examining the brain tissue of an individual post-mortem. A neuropathological diagnosis of CTE is currently available and is widely accepted and recognized in the medical and scientific community as a valid diagnosis. The neuropathology associated with CTE is unique and distinct from the neuropathology of other

neurodegenerative diseases. As I wrote, “in the 21st century, the challenge is no longer the acceptance of the entity of CTE but rather a sorely needed accounting of the actual numbers of affected persons as well as the numbers of those who remain unaffected despite exposure to the identical repetitive head traumas.” Ex. A at 2.

10. Second, although there currently are no *in vivo* biomarkers that can be used to reliably diagnose CTE, “PET imaging ligands are now in development for the detection of abnormally phosphorylated tau associated with [the] neurofibrillary tangles” that are characteristic of CTE. Ex. A at 16. A PET imaging ligand is a molecule that binds to a certain protein in the body, in this case phosphorylated tau. The ligand is detectable using a PET scan and thus allows physicians and researchers to image the pattern and location of the protein in the body. Indeed, recent reports suggest that one such PET imaging ligand—T807—has been successfully employed for the detection of CTE tauopathy *in vitro* and *in vivo*. Ex. A at 17. It is only a matter of time before this ligand, and others like it, will be sufficiently developed that they can be used in conjunction with clinical diagnostic criteria to render a reliable clinical diagnosis of CTE in the living.

11. I have reviewed the declarations of Drs. Bernick, DeKosky, Hof, Shenton, Stone, Weiner, Wisniewski, and Zhang. Each of those experts stated that a “clinically accepted diagnosis of CTE” will likely be possible within ten years and will be possible before the expiration of the Settlement term. I understand – as would any expert in the field, including the declarants named above – that a “clinically accepted diagnosis” is a diagnosis in a living person.

12. Mr. Seeger also attributed to me the remark that “we have little idea, however, of the risk of developing CTE following [traumatic] brain injury.” *See* Tr. 196:11-16. That quote is taken from an article on which I was listed as a co-author. The article appeared in in the

journal *Nature Review Neurology* and is titled “Acute and Chronic Traumatic Encephalopathies: Pathogenesis and Biomarkers.” That article is attached as Exhibit B. Nothing in that article undermines or contradicts the statements in my October 14 declaration.

13. Again, Mr. Seeger takes this quote out of context and ignores what *is known* about CTE.

14. Repetitive brain trauma is a *necessary condition* for developing CTE; in the absence of repetitive brain trauma, an individual will not develop CTE. There is “definitive documentation that progressive neurodegeneration . . . was associated with elective exposure to repetitive head trauma.” Ex. A at 1. And in nearly 200 cases of neuropathologically confirmed CTE, *every case* has occurred in an individual who experienced repetitive brain trauma. Indeed, CTE has not yet been found outside the population of individuals exposed to repetitive brain trauma.

15. There is also an “increasingly evident association of CTE with American football” and the disease “has also been associated with other high impact sports (soccer, hockey) and with exposure to improvised explosive devices (IEDs) in the battlefields of Iraq and Afghanistan.” Ex. A at 2; Ex. B at 5. In fact, I find this association sufficiently strong that I believe informed consent protocols should be developed and required of individuals contemplating exposure to repetitive TBI.

Pursuant to 28 U.S.C. § 1746, I state under penalty of perjury that the foregoing is true and correct.

Dated: December 2, 2014

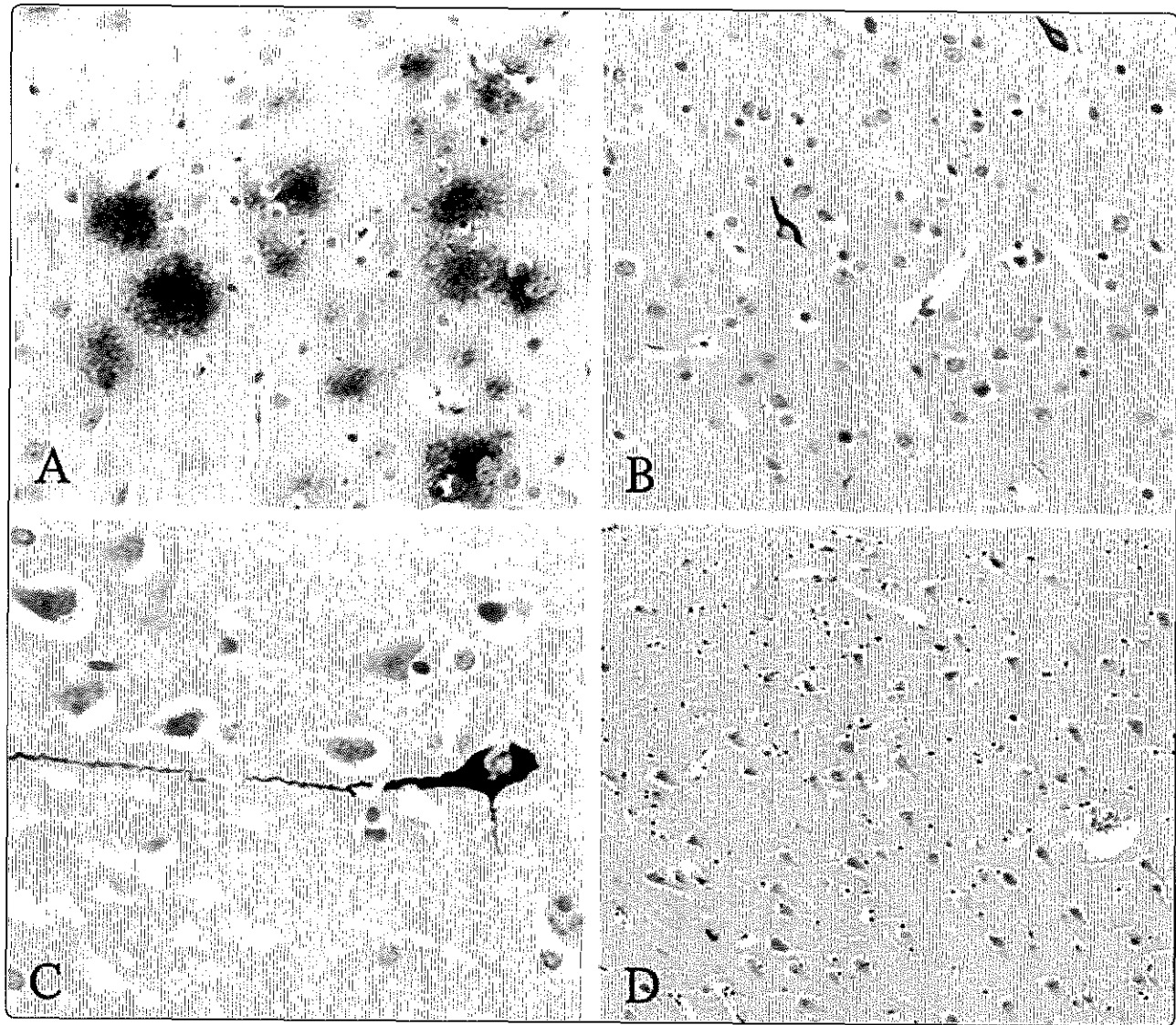
Sam Gandy, M.D., Ph.D.

Sam Gandy, M.D., Ph.D.

SAM GANDY MD PhD

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Exhibit A



Chronic traumatic encephalopathy: clinical-biomarker correlations and current concepts in pathogenesis

Gandy *et al.*

REVIEW

Open Access

Chronic traumatic encephalopathy: clinical-biomarker correlations and current concepts in pathogenesis

Sam Gandy^{1,2,3,4*}, Milos D Ikonovic⁵, Effie Mitsis^{2,3*}, Gregory Elder^{1,2,3,4}, Stephen T Ahlers⁶, Jeffrey Barth⁷, James R Stone⁸ and Steven T DeKosky⁹

Abstract

Background: Chronic traumatic encephalopathy (CTE) is a recently revived term used to describe a neurodegenerative process that occurs as a long term complication of repetitive mild traumatic brain injury (TBI). Corsellis provided one of the classic descriptions of CTE in boxers under the name "dementia pugilistica" (DP). Much recent attention has been drawn to the apparent association of CTE with contact sports (football, soccer, hockey) and with frequent battlefield exposure to blast waves generated by improvised explosive devices (IEDs). Recently, a promising serum biomarker has been identified by measurement of serum levels of the neuronal microtubule associated protein tau. New positron emission tomography (PET) ligands (e.g., [¹⁸F] T807) that identify brain tauopathy have been successfully deployed for the *in vitro* and *in vivo* detection of presumptive tauopathy in the brains of subjects with clinically probable CTE.

Methods: Major academic and lay publications on DP/CTE were reviewed beginning with the 1928 paper describing the initial use of the term CTE by Martland.

Results: The major current concepts in the neurological, psychiatric, neuropsychological, neuroimaging, and body fluid biomarker science of DP/CTE have been summarized. Newer achievements, such as serum tau and [¹⁸F] T807 tauopathy imaging, are also introduced and their significance has been explained.

Conclusion: Recent advances in the science of DP/CTE hold promise for elucidating a long sought accurate determination of the true prevalence of CTE. This information holds potentially important public health implications for estimating the risk of contact sports in inflicting permanent and/or progressive brain damage on children, adolescents, and adults.

Overview

Chronic traumatic encephalopathy (CTE) is unique among brain diseases in having a history of decades of organized opposition to its codification as an authentic or valid entity. The conceptual entity has evolved over the 75 years since Harrison Stanford Martland [1], writing in *The Journal of the American Medical Association* in 1928, coined the term "punch drunk" to describe the tremors and

impaired cognition that affected some boxers [1]. In 1937, Millspaugh [2] coined the term "dementia pugilistica" (DP), which was broadened to "chronic traumatic encephalopathy" (CTE) by Macdonald Critchley [3] in 1949. In 1973, John Corsellis and colleagues [4] brought DP/CTE into the modern day with their definitive documentation that progressive neurodegeneration (Figures 1 and 2) was associated with elective exposure to repetitive head trauma. This report set off a controversy that continues today regarding the role of society in regulating intentional head injury and in establishing the liability of organizations that encourage such exposure with little in the way of informed consent.

Despite condemnation of boxing by the American Medical Association [6] and by the American Pediatrics

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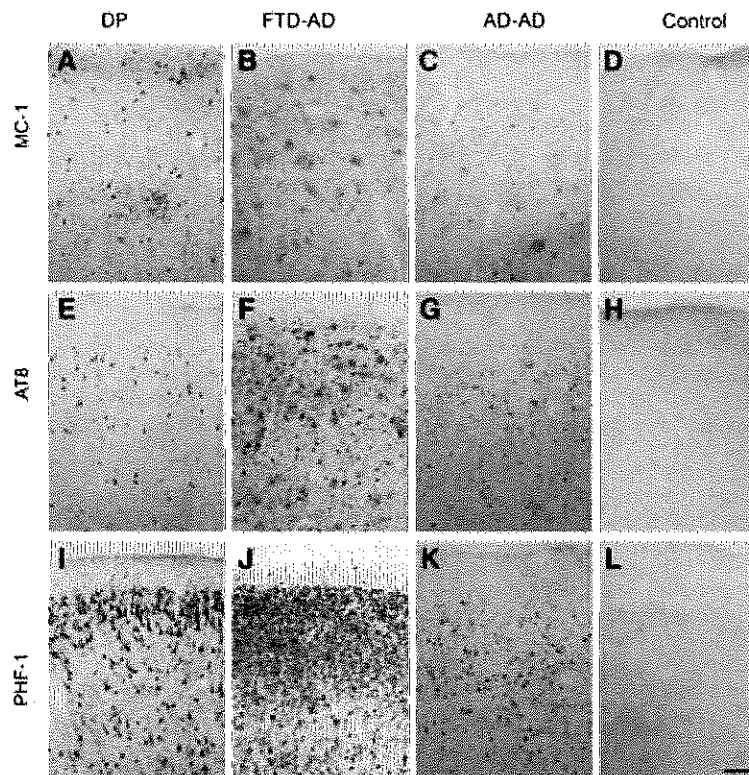


Figure 1 Patterns of tau immunostaining in the frontal cortex of the patient with dementia pugilistica (DP), compared to Alzheimer's disease (AD) and nondemented cases. MC-1 immunolabeling of phosphorylated tau revealed neuronal labeling in the DP case (A), plaque-associated neuritic labeling in the frontotemporal dementia (FTD)-AD case (B), a mix of neurofibrillary tangles (NFTs) and dystrophic neurites in the typical AD case (C), but absent in the control case (D). AT8 immunoreactivity also was limited to intracellular NFTs in the DP case (E), and within NFTs and plaque-associated dystrophic neurites in the FTD-AD case (F) and the typical AD case (G), but were not present in the control case (H). PHF-1 immunostaining was the most extensive of the three pathological tau markers, and showed significant intracellular and extracellular NFT labeling in the DP case (I), and within NFTs and plaque-associated dystrophic neurites in the FTD-AD case (J), but was less extensive and limited to NFTs in the typical AD case (K). No PHF-1-positive tangles were observed in the control case (L; scale bar = 100 μ m). From Saing et al. [5] with permission.

Association [7] in 1997 and predictions of the rapid demise of boxing, the sport continues today, and multi-million dollar purses still await the winners. In turn, the chance to compete for these purses continues to attract new boxers into professional associations, fueling the sport. Other sports associated with repetitive head trauma (e.g., ultimate fighting and mixed martial arts) have expanded and found larger participant groups and fans. Despite much social opposition to these sports, all efforts at boxing bans have so far been stymied by the influence of pro-boxing lobbyists on state and federal legislatures [8]. While over 50 sports-associated cases of CTE have been reported [9], efforts at educating the public regarding the risks of repetitive head injury are hindered by some retired boxing champions who refuse to recognize that their own brain disease is due to their chronic exposure to boxing. Newer "sports" involving various forms of fighting provide further encouragement.

In 2005, Omalu and colleagues [10] revived the term CTE in their report of the index case of a retired

National Football (NFL) player with progressive neurological dysfunction (Figure 3). The term CTE includes DP and supplants the use of the term DP. With the increasingly evident association of CTE with American football, the stakes grew exponentially. In part as a result of the NFL's consistent denial of the danger of CTE to its players, in both public statements and legal challenge statements, the disease remained out of the spotlight until the League aligned itself with the independent and academic experts who were studying the disease [11]. CTE has also been associated with other high impact sports (soccer, hockey) and with exposure to improvised explosive devices (IEDs) in the battlefields of Iraq and Afghanistan [12-14] (Figure 4). Now, in the 21st century, the challenge is no longer the acceptance of the entity of CTE but rather a sorely needed accounting of the actual numbers of affected persons as well as the numbers of those who remain unaffected despite exposure to the identical repetitive head traumas. The role of aging has also gone unexplored. With those numbers in hand, we

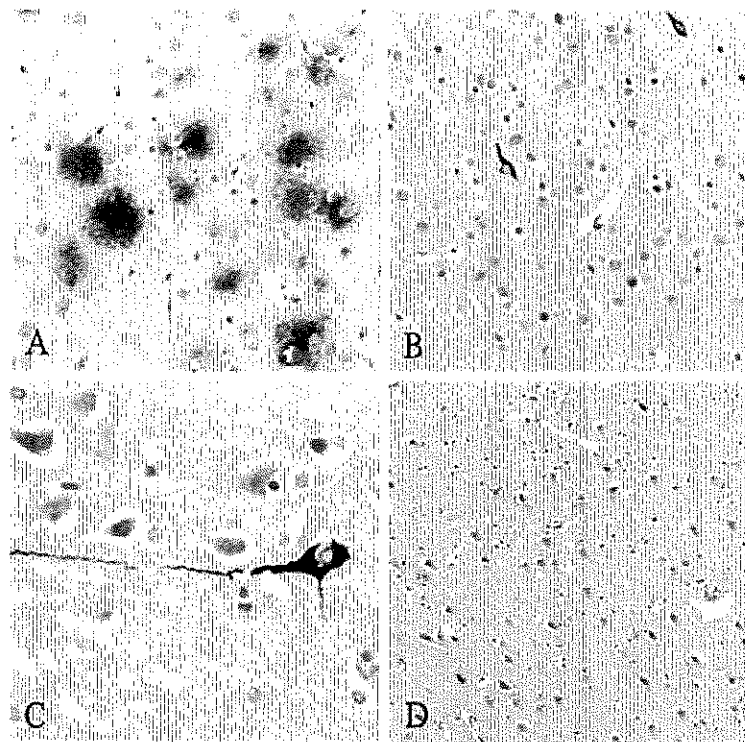


Figure 3 Micrographs from the index case of CTE in an American football player. Panel A, β -amyloid immunostain of the neocortex (original magnification, $\times 200$) showing frequent diffuse amyloid plaques. Panel B, tau immunostain of the neocortex (original magnification, $\times 200$) showing sparse NFTs and many tau-positive neuritic threads. Panel C, tau immunostain (original magnification, $\times 400$) showing an NFT in a neocortical neuron with extending tau-positive dendritic processes. Panel D, β -amyloid immunostain (original magnification, $\times 100$) of the Sommer's sector (CA-1 region of the hippocampus) showing no diffuse amyloid plaques. From Omalu *et al.* [10] with permission.

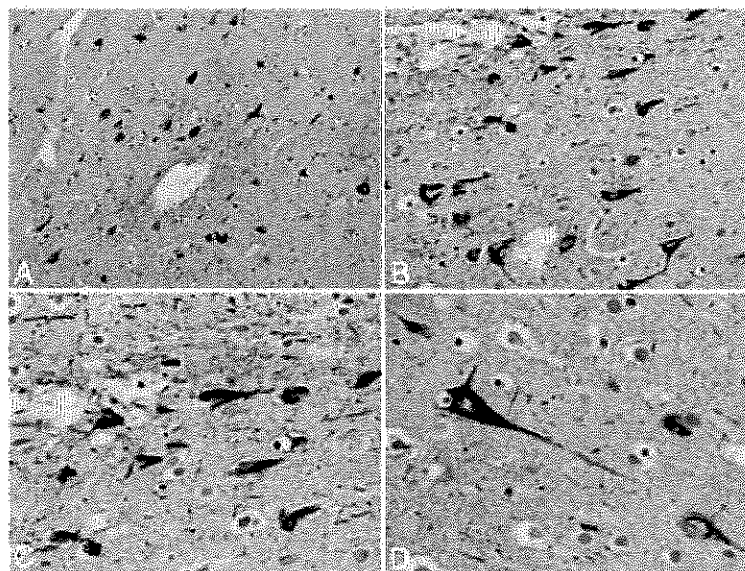


Figure 4 The index case of military CTE. Photomicrographs of tau-immunostained section of the frontal cortex showing frequent neurofibrillary tangles and neuritic threads (A and B), with higher magnification (C and D) showing band- and flame-shaped neurofibrillary tangles and neuropil neuritic threads. Original magnification $\times 200$ (A), $\times 400$ (B), $\times 600$ (C and D). From Omalu *et al.* [15] with permission.

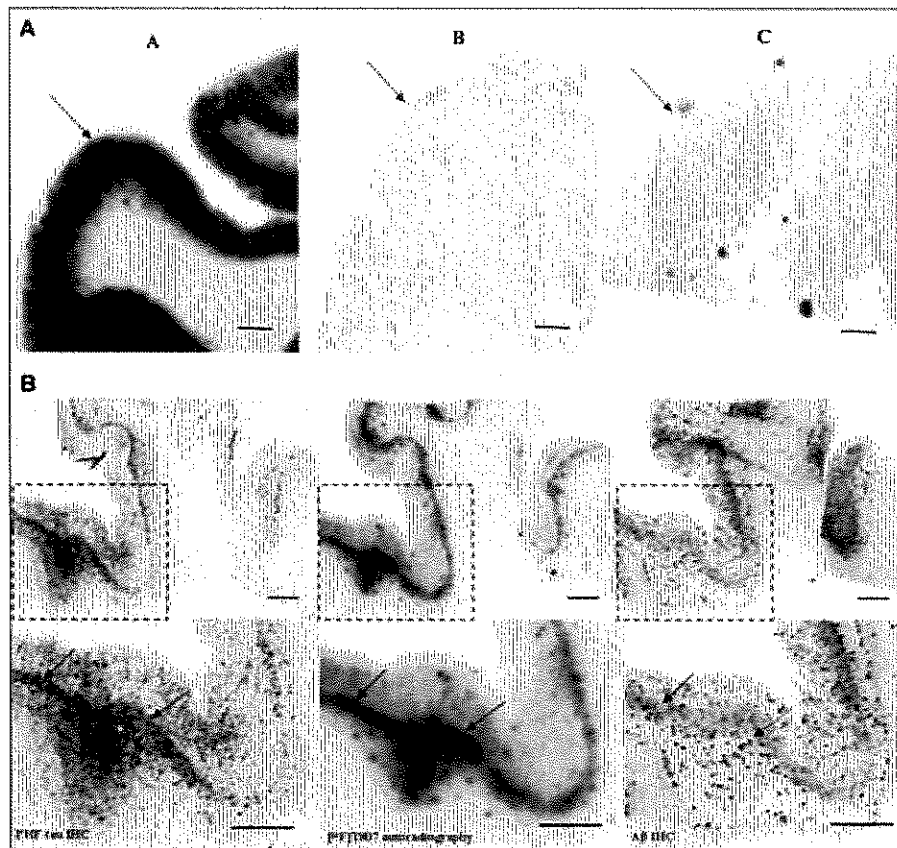


Figure 5 [18 F] T807 autoradiography on brain sections and its comparison with paired helical filament (PHF)-tau and amyloid beta (Aβ) double immunohistochemistry (IHC). (A) Representative images for [18 F] T807 autoradiographs from groups A, B, and C of brains ([18 F] T807 autoradiography, 20 μ Ci/section). Positive autoradiography signals were observed only in the gray matter of brain from the PHF tau rich group A. Arrows indicate gray matter. (B) [18 F] T807 colocalized with PHF-tau but not with Aβ plaques. (B, top row) Low magnification. (B, bottom row) High magnification from the framed areas. Images of PHF tau (left) and Aβ (right) IHC double immunostaining and autoradiogram image (middle) from two adjacent sections (10 μ m) from a PHF-tau rich group A brain (frontal lobe). Positive [18 F] T807 labeling colocalized with immunostaining of PHF tau but not with Aβ plaques, as indicated by arrows. Fluorescent and autoradiographic images were obtained using a Fuji Film FLA-7000 imaging instrument. Scale bars = 2 mm. From Xia et al. [19] with permission.

from the study of boxers and, more recently, football players, athletes from other contact sports, as well as those exposed to domestic violence, and military personnel exposed to battlefield blast injuries [20,21]. However, there have been no prospective studies linking clinical phenotypes during life (including neuropsychological function and outcome) with autopsy-confirmed CTE. Thus, the clinical and neuropsychological characterization of CTE is yet to be properly developed. The development of biomarkers for CTE (reviewed in detail below) should greatly facilitate the fulfillment of this goal.

In their review of 48 cases of neuropathologically confirmed CTE, McKee et al. [21] found that memory loss was reported in over half of the individuals. As in AD, loss of insight often precluded patients from recognizing their deficits, and this important piece of the presentation was derived from friends or family. Other studies have reported that impairment in executive function is common

in neuropathologically confirmed CTE [22]. Executive functions are a collective set of higher order abilities (judgment, self-inhibitory behaviors, decision-making, planning and organization) considered to be primarily dependent upon adequate functioning of the frontal lobes of the brain. Damage to various regions of the frontal cortex can disrupt these higher order abilities, leading to poor impulse control, and socially inappropriate, avolitional, and apathetic behaviors. For example, damage to the orbitofrontal regions can result in significant changes in personality. Thus, changes in personality, apathy, impulsivity, aggression, and "short fuse" behaviors typical of CTE [23] are consistent with the atrophy and other neuropathological changes of the frontal lobes that have been described in nearly all reported cases of CTE [16,21-24].

Neuropsychological, mood, and neurobehavioral dysfunction in CTE typically presents in midlife after a latency period, usually years or decades after exposure to the

repetitive trauma. The cognitive and behavioral symptoms of CTE begin insidiously, followed by progressive and gradual deterioration. Mood symptoms are typically depression, apathy, irritability, and suicidality. Behavior symptoms are impulse control, disinhibition, and aggression as well as comorbid substance abuse [24]. The continued degeneration of brain regions most severely affected in CTE (cerebral cortices, hippocampi, amygdalae, basal forebrain, mammillary bodies) results in the worsening of behavioral and mood symptoms, and the further deterioration of cognitive abilities subserved by these regions. Baugh *et al.* [24] organized the neuropsychological and neuropsychiatric symptoms of CTE into the categories of cognition, mood, and behavior. As mentioned, the early cognitive symptoms of CTE involve memory impairment and executive dysfunction. The early involvement of hippocampal-entorhinal cortices and medial thalamic circuits may explain the memory impairment and its similarity to the memory loss associated with AD. As in AD, the genetic risk posed by apolipoprotein E $\epsilon 4$ (*APOE* $\epsilon 4$) may play a role in the dementia associated with CTE [25-27]. The dysexecutive syndrome in CTE may result from the early neurofibrillary degeneration of the frontal cortex [10,21,22]. As the disease progresses, there is worsening memory impairment and executive dysfunction, language problems, motor dysfunction, aggression (physical as well as verbal), and apathy [23]. Dementia is evident in most CTE individuals who live to be over the age of 65 years [24]. However, the high rates of suicides, accidents, and drug overdoses often lead to death prior to this age [10,28]. As a result, most persons with neuropathologically confirmed CTE were not demented at the time of death. In view of the young age of onset as compared with AD, CTE may be misdiagnosed as the behavioral variant of FTD. However, CTE has a more gradual and prolonged progression than does FTD, and CTE is associated with a repetitive TBI history but with no family history [24].

Neuropsychological testing may be valuable in scientific studies on TBI in both the acute and chronic phases. Athletes and military combatants exposed to multiple blast events are at highest risk for sustaining multiple mild TBI or concussions and potentially developing CTE. In order to appreciate fully the effects of single and multiple concussive and sub-concussive brain injuries and to enable tracking of recovery in individual cases, brief baseline and serial post-concussion neurocognitive assessments, based upon the Sports as a Laboratory Assessment Model (SLAM) methodology, are the recommended standard of practice [29]. Key neurocognitive elements of these abbreviated assessments include attention, processing speed, reaction time, and learning and memory.

Once CTE is suspected, based upon a history of repeated trauma and presence of cognitive and/or behavioral impair-

ment, scientific studies are needed to evaluate the value of comprehensive neuropsychological testing; e.g., assessment of general verbal and visuospatial problem solving, language fluency, attention, learning and memory, speed of mental processing, abstract reasoning, judgment, new problem solving, planning and organization, mental flexibility, sensory and motor intactness, and emotional/psychological and behavioral functioning. A recent cross-sectional study assessed cognitive impairment and depression, as well as the neuroimaging correlates of these dysfunctions, in former NFL players [30]. Former NFL players with cognitive impairment and depression were compared to cognitively normal retired players who were not depressed, and a group of matched, healthy control subjects. The cognitive deficits found were primarily in naming, word-finding, and memory (verbal and visual) and were associated with disrupted white matter integrity on DTI and changes in regional cerebral blood flow. Although none of the players fit the clinical profile of CTE *per se*, the sample size was very small. Nevertheless, this is one of the first studies examining the neural correlates of cognitive dysfunction and depression in players with a history of concussions (range 1-13 concussions in this sample).

Neuropathological changes in boxers (dementia pugilistica, punch-drunk syndrome)

For this section, the combined term DP/CTE is employed, since the term DP appears in this original literature. However, the revival of the term CTE was intended to include DP and to supplant the use of the term DP. In 1934, Parker described three cases of boxers affected with the illness [31] and drew attention to the risk of developing DP/CTE in professional boxing. Neuropathological examination of these retired professional boxers' brains demonstrated that the primary DP/CTE lesion involved multifocal intracellular aggregates of hyperphosphorylated tau, which resembled the neurofibrillary tangles (NFT) found in AD brains [32]. Using Congo red and silver staining, Corsellis and colleagues studied 15 cases of DP/CTE and confirmed the abundance of NFT. Like the NFT of AD, the NFT of DP/CTE is also reactive to antibodies to other AD-related proteins such as ubiquitin [33] and A β [34]. The anatomical patterns of NFT distribution are different in DP/CTE and AD: there is greater involvement of superficial layers of the associational neocortex in cases with DP/CTE and notably in the depths of cortical sulci [35]. This difference may reflect a unique way in which NFT are formed and propagated in DP/CTE.

In clear distinction to neuropathologically confirmed AD, no congophilic or argyrophilic plaques were observed in the Corsellis study [32]. In 1991, other investigators re-examined 14 out of the original 15 DP/CTE cases from the Corsellis study as well as additional brains of professional and amateur boxers, using A β immunohistochemistry and formic acid pre-treatment of tissue

sections [34,36-38]. Re-examination revealed that these 14 brains with NFT pathology (and even two amateur fighters' brains that lacked NFT) also had A β -immunoreactive plaques [34,36-38]. These DP/CTE plaques were described as diffuse in most cases; they lacked both Congo red positivity and the silver-positive dystrophic neurites that are typical of mature AD plaques. Some cases also displayed cerebrovascular deposits of A β . These data demonstrated that DP/CTE is associated with both the NFT and A β -related AD-like pathology. The antigenic similarities of neurofibrillary lesions in DP and AD led Roberts to suggest that the two conditions likely share the same pathogenesis and that TBI may be a risk factor for developing AD [37].

More recent studies reconfirmed that DP/CTE includes both NFT and A β pathology. Tokuda *et al.* [38] and Nowak *et al.* [39] examined the brains of boxers who suffered a progressive cognitive decline before death, and reported NFT as well as infrequent neuritic A β plaques. McKee and colleagues [21] studied two aged (80 and 73 years) professional boxers and presented them along with 37 previously published cases of boxers with neuropathologically verified DP/CTE. Similar to earlier reports, the most striking pathological feature in these two cases involved multifocal patches of neuronal and astrocytic NFT in superficial cortical layers, most frequently at the depths of the sulci and around large blood vessels [21]. In one of the two boxers, there were moderate diffuse A β plaques and sparse neuritic plaques in several neocortical regions. Collectively, these reports indicate that the neuropathology of DP/CTE was heterogeneous and involves both NFT and diffuse A β deposits, although the extent and proportions of these lesions vary from case to case and may be influenced by other factors such as number of years since TBI, age at the time of the TBI, and genetic predisposition. In a study by Geddes and colleagues, the brain of a 23-year-old boxer was found to have all neocortical areas affected with NFT; however no other changes (including A β deposits) were reported [40]. APOE $\epsilon 4$ alleles have been associated with more severe cognitive deficits in boxers [25] and could contribute to more severe A β pathology, as has been demonstrated in AD. In an APOE $\epsilon 4$ heterozygous retired boxer with DP/CTE, death was caused by hemorrhage due to amyloid angiopathy [41]. Clinical misdiagnosis of DP/CTE as AD is not unusual; interestingly, both boxers in the McKee report were diagnosed with AD during life. Differential diagnosis is likely to be clarified with amyloid imaging. We reported a case report illustrating this wherein the clinical diagnosis of retired NFL player could not be resolved until florbetapir imaging excluded the diagnosis of AD [42].

Other common findings in DP/CTE include cerebral infarcts and fenestrated cavum septum pellucidum (CSP) as well as substantia nigra degeneration [28]. Changes in substantia nigra could explain the increased incidence of

Parkinsonism in retired boxers. It had been proposed that identification of CSP on CT scans could be of a diagnostic value for DP/CTE [43]. However this pathological feature is not consistently present [44], possibly because of the high prevalence of CSP in both boxers and non-boxers [45]. Because of the rigid enclosure of the brain inside the calvarium, acceleration-deceleration injuries associated with boxing often involve cerebral and meningeal vascular damage, the most common of which is subdural hematoma and to a lesser extent epidural, subarachnoid, and/or intraparenchymal hemorrhages [46]. Neuropathological sequelae of boxing appear to be less severe in amateur boxers, likely due to better-implemented regulations, fewer total bouts, and other measures of protection. In retired professional boxers, however, the risk of developing DP/CTE is greater: the extent of neurological abnormalities defined by CT and EEG, and severity of DP/CTE symptoms, correlated with numbers of fights and overall duration of boxers' career [25,47,48].

Not shown here are some other CTE-associated structures described by McKee [21] including astrocytic tangles, perivascular tau pathology, or patchy tau pathology. An important significance for these structures is that McKee uses their presence to differentiate CTE from AD. The reader is referred to [21] for typical images of those structures.

Neuropathological changes associated with American football

In contrast to the neurodegenerative changes associated with professional boxing, the chronic neuropathological sequelae of repetitive hits to the head in American football and other contact sports (hockey, rugby, etc.) have only been recognized more recently. Impacts to the head in American football cause less rotational acceleration than those suffered in boxing [49]; football-related injuries are of a translational acceleration-deceleration type, and despite the use of helmets, they can result in concussions or unconsciousness. In 2005, Omalu and colleagues reported the first neuropathology finding of CTE in a 50-year old retired NFL player [10]. In this case, they described AD-like changes that consisted of neocortical A β -immunoreactive plaques and sparse tau-immunoreactive neuronal and axonal aggregates resembling NFT and neuropil threads. While coexistence of A β plaques and NFT might have suggested an ongoing AD process, several observations argued against this idea: 1) there was no family history of AD; 2) at autopsy, the brain had no signs of cortical atrophy or overt neuronal loss; 3) the neocortical A β -immunoreactive plaques were numerous, but were diffuse (non-neuritic); and 4) NFT were scarce in the neocortex and absent in the entorhinal cortex and hippocampus which is the initial focus of NFT development in AD, prior to any cortical NFT [50]. The neuropathological pattern consisting of diffuse neocortical A β plaques and scarce

NFT was similar to changes described in subjects with acute severe TBI [51] or in cases with preclinical AD, suggesting that both mild repetitive and severe brain injuries may initiate early AD-like changes. The second case reported by Omalu and colleagues [52] was a 45-year old retired NFL player who had numerous cortical and subcortical tau-immunoreactive NFT and neuropil threads, but no A β -immunoreactive plaques. In a more recent study, McKee and colleagues [21] described CTE in a 45-year old retired football player and compared their findings to previous neuropathology reports in four players who had played similar positions during their career. Tau-immunoreactive NFT-like neuronal and glial (fibrillar astrocytic) profiles and thread-like neuropil neurites were frequent in multiple cortical regions, predominantly occupying deep sulcal and perivascular locations [21]. NFT were also abundant in the entorhinal cortex, hippocampus, amygdala, nucleus basalis and septal nuclei, hypothalamus, thalamus, striatum, and olfactory bulb. Diffuse A β plaques were reported in 3 of the 5 cases [21]. As in CTE due to boxing (i.e., DP), there are similarities of the effects of football injuries to AD and other neurodegenerative disorders, notably involving NFT and diffuse A β deposition, and these findings require further investigation.

Neuropathological changes associated with battlefield blast exposure

Studies of military servicemen who participated in recent wars indicate that repetitive mild head injury and concussions due to blast (explosive) injury might also result in CTE. This literature is the least mature since the index case of military CTE was only reported by Omalu a few years ago [15]. Brody and colleagues used diffusion tensor imaging (DTI) to evaluate the extent of axonal injury in 63 U.S. military personnel diagnosed with mild TBI after blast exposure with secondary mechanical injuries [53]. They found that, in these subjects, TBI was associated with significant axonal injury, which was still present 6–12 months later at follow up evaluation with DTI. Diffuse loss of white matter integrity was also detected using high angular resolution diffusion imaging (HARDI) in military veterans from Iraq and Afghanistan who had been diagnosed with mild TBI and comorbid PTSD [54]. Collectively, these findings indicate that TBI associated with blast or chronic mild injury produces chronic neuronal damage. Blast injuries and closed head injuries have similar clinical features [55], however, the extent of neurological damage in blast injured veterans is likely influenced by injuries of other systems, since air-filled organs (including lungs) are often severely affected [56,57].

Goldstein [58] and colleagues performed postmortem analyses of brain tissues from four military veterans who were exposed to blast or concussive injury and compared them to four professional athletes who suffered repetitive

concussions. In the neocortex from military veterans, there were frequent perivascular and deep sulcal accumulations of NFT-like tau-positive neurons and glial cells as well as dystrophic axons were reported [58]. Subcortical white matter in close proximity to these lesions had dystrophic axonal changes and clusters of activated microglia were described. These pathological changes were similar to those observed in sports CTE. The findings in this study have been challenged because all military CTE subjects had histories of *both* civilian and military TBI, making it impossible to distinguish which lesions arose from which injuries. Similar tau-immunoreactive neuropathological features were described in an Iraqi war veteran with PTSD who had been exposed to multiple blast explosions during his deployments [15]. These autopsy studies of brains from blast-injured veterans are not in agreement with recent body fluid biomarker analyses in soldiers exposed to blast [53], emphasizing both the variability of such injuries and the variability in the accuracy of the historical details surrounding the injuries. In this study of Army officers exposed to different levels of blast overpressure by firing heavy weapons, Blennow and colleagues [59] measured CSF biomarkers of neuronal injury (tau and neurofilament protein) or glial injury (GFAP and S-100 β) as well as CSF/serum albumin ratio, hemoglobin, and bilirubin content in CSF. GFAP and S-100 β were also analyzed in serum samples. All analyses indicated normal levels of examined markers, leading the authors to conclude that high-impact blast was not associated with biomarker evidence of brain damage [59]. The discrepancy between this report and published evidence of neuropathology in blast injured servicemen warrants further investigation, and the issue of whether the blast wave itself causes injury remains open. Another possible explanation is the lag time between injury and serum sampling. Both [53] and [59] illustrate the discrepancy between neuropathology that can be static and long-lasting vs serum biomarkers that will rise acutely and then fall as the marker molecule is cleared. Recent studies of serum tau as a marker of acute TBI in hockey players showed surprisingly good correlation with outcome [60].

Molecular pathogenesis of CTE

As noted above, CTE is considered to be primarily a tauopathy with frequent coexistence of A β pathology. Notably acute severe TBI is associated with diffuse A β plaques and minimal tau pathology [51]. Other types of abnormal protein aggregates are less common in CTE; for example, alpha-synuclein immunoreactive inclusions were not reported in any of 51 CTE cases reported by McKee and colleagues [21]; however, they were detected only following acute severe TBI [51]. Multiple cases of Parkinsonism have been reported in professional boxers, and there is increased prevalence of ALS in professional football [61] and soccer players [62].

The detailed molecular pathogenesis of CTE is unknown. As observed in cases with single severe head trauma, axonal injury occurs in CTE, and this may be responsible for accumulation of phosphorylated tau. Tau is a microtubule-associate protein which can undergo excessive phosphorylation in the pathological milieu of brain injury associated with ischemic foci that increase the risk of damage from oxidative stress. The idea of ischemic changes as a contributing factor to tau hyperphosphorylation and formation of NFT-like changes in CTE is consistent with their preferential deep sulcal and perivascular localization. Because both neuronal and astrocytic NFT are observed in these areas, it is possible that changes in tau are related to a more general injury response mechanism that is not injury-specific or cell-specific (e.g., brain ischemia, inflammatory reaction).

Diffuse axonal injury (DAI) is the most consistently reported neuropathological feature following acute head trauma [63]. DAI is associated with intra-axonal accumulation of tau, A β -precursor protein (APP), ubiquitin, and α -synuclein [64,65]. As an acute reaction to TBI, there is an upregulation of APP and/or a switch in its metabolic processing resulting in increased concentrations of A β , which can deposit in diffuse, predominantly A β 42, plaques both acutely after severe TBI [51] and chronically in boxers with DP/CTE. In studies of both autopsy cases and in surgical biopsy tissue from subjects with TBI [51], about 30% of acute severe TBI patients are reported to show evidence of A β plaques. A β deposits in CTE have received less attention in the literature than has the tauopathy; however these changes may have a significant role. The development of A β lesions may depend on age of the subject, time interval since the head injury, and genetic factors. More extensive and mature A β pathology (i.e., neuritic plaques) may be absent, or may recede over years, in subjects with protective genetic factors that can result in better degradation/clearance of A β from the brain [66,67], while tau-related pathology may develop more slowly, may be more resistant to degradation and therefore observed at late stages of disease. Other genetic factors (*APOE4*) may influence A β pathology. The link of *APOE4* to dementia in CTE is unexpected on the basis of tauopathy, since *APOE4* modulates A β pathology in AD but not tauopathy. Perhaps *APOE4* modulates the severity of the acute A β deposition which, in turn, affects clinical severity. This is a key point in determining which types of interventions will be most promising and at what point in pathogenesis each is most likely to be successful. Recent evidence that chronic action of interleukin-1 β can reduce amyloidosis while exacerbating tauopathy raises the possibility that this neurodegeneration-associated cytokine may drive the conversion of acute post-traumatic A β deposition to chronic tauopathy with only minimal or inconsistent A β residua [68].

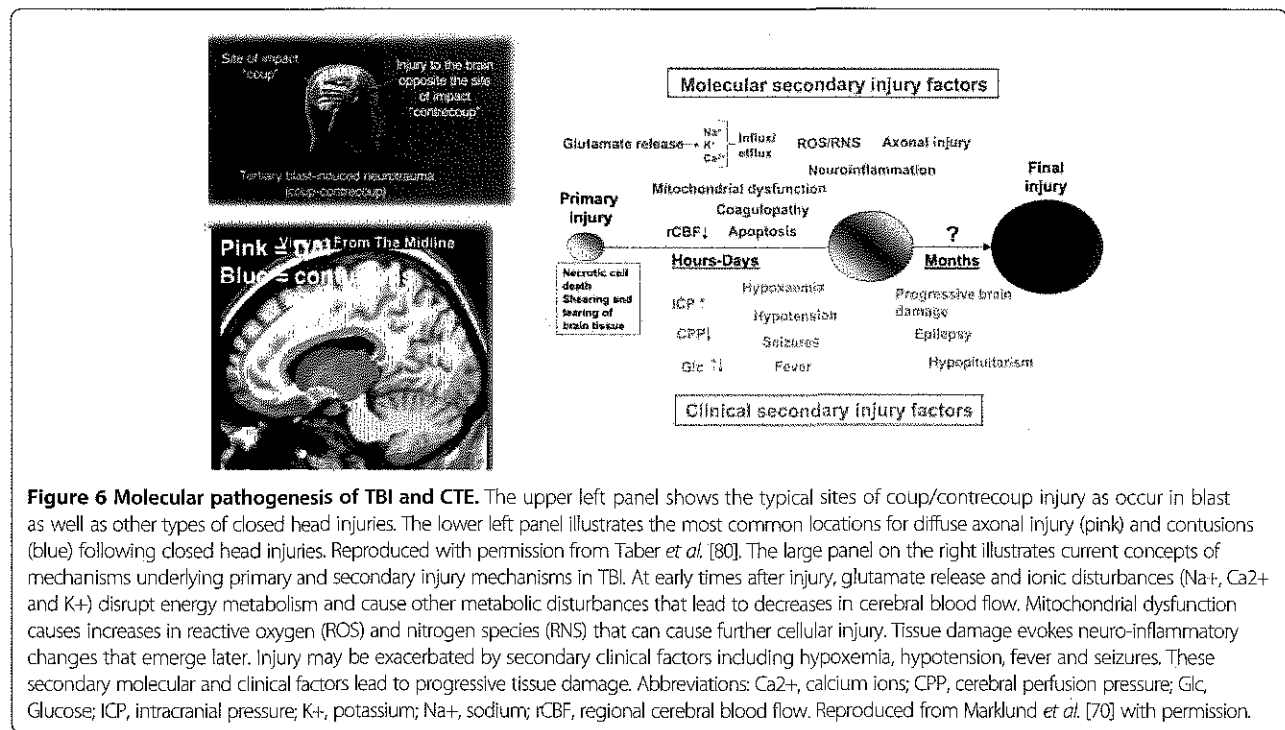
Alternatively, the extent and type of neuropathology changes in CTE resulting from different contact sports may reflect differences in the biomechanics associated with head injury, including impact level (force) and type of head movement (rotation, acceleration, deceleration). Novel genetic, biomarker, and neuroimaging analyses should be developed and employed for early detection of neuropathology, to provide a window of opportunity for preventing or at least delaying the clinical manifestations of neurodegenerative disease after TBI. Furthermore, timely identification of people at risk (e.g., *APOE4* carriers) [27], and regular testing (imaging, CSF analyses, and detailed neuropsychological evaluation) should be considered essential.

Modeling CTE in laboratory animals

Currently there is no experimental animal model that recapitulates all pathophysiological aspects of human CTE. The best understood pathophysiological mechanisms associated acutely with severe blunt impact TBI are hemorrhage, mechanical tissue damage, and diffuse axonal injury (DAI) [69]. DAI results when angular forces cause shearing or stretching of axons; this can impair axonal transport which is manifested pathologically by focal axonal swellings, most commonly at gray/white matter junctions particularly in frontal and temporal cortical regions. Contusions occur as the result of coup/contrecoup injuries most commonly affecting the frontotemporal and occipital cortex. However not all injury occurs at the time of initial impact. Rather a complex cascade of pathophysiological effects unfolds over the ensuing hours and days following injury and results in further tissue damage [70] (Figure 6). Factors thought important in this secondary injury cascade include release of excitatory amino acids such as glutamate, calcium dyshomeostasis, mitochondrial dysfunction and oxidative stress. Increased glucose utilization at a time of decreased cerebral blood flow also likely exacerbates injury. While these molecular mechanisms are thought to be operative in moderate to severe TBI, there is much less in the way of neuropathological or molecular data on mild TBI and particularly from the mild repetitive concussions that contribute to CTE. What sets CTE apart from other injuries is a relentlessly progressive course leading to a syndrome that continues to progress even in the absence of further head trauma. Thorough understanding of this pathological transition in CTE would be facilitated greatly by an appropriate animal model.

Relevance of existing animal models of TBI to human CTE

Because of its potential clinical importance, animal modeling of TBI has been vigorously pursued and a number of methods have been developed that can induce focal, diffuse or mixed brain injury [70]. Most studies of TBI use rodents, but rabbits, pigs, cats, dogs, and nonhuman



primates have all been studied as well [69]. The choice of species has practical as well as theoretical implications. While rodents are less expensive than larger animals, their lissencephalic brains lack the structure of gyri and sulci found in humans. The white/grey matter ratio is also less in rodents compared to human brain; these anatomical factors may affect the biophysical properties of the brain's reaction to mechanical injury. Species such as pigs and non-human primates offer the advantage of having a brain more similar to humans, but their cost and in the case of non-human primates availability limit their wider use for TBI research.

To model various types of TBI in experimental animals, several methods have been developed [70]. Among the more widely used models, controlled cortical impact (CCI) produces focal contusions with pericontusional axonal injury while fluid percussion models produce more diffuse axonal injury. Weight drop methods can produce a range of injuries depending on the force applied (mass x distance of the drop), and whether an open skull or closed skull technique is used. There has been recently increased interest in models of blast injury, due primarily to its importance in military head trauma [71]. Importantly, these models are associated with significant deficits in cognitive and motor function. In addition, anxiety and depression are common features of the post-concussion syndrome and evidence for impairment in both domains have been found as sequelae of TBI in some animal models [72]. There are several recognized limitations of the animal models. While human TBI

represents a heterogeneous injury, most animal models try to replicate more isolated pathological factors [69]. Human TBI is frequently accompanied by hypoxia, hypotension and ischemia, factors that are not prominent in many animal models [69]. Accordingly, TBI models can be utilized in combination with additional insults such as hemorrhagic shock which often accompanies blast TBI [71]. Another limitation of traditional TBI models in replicating CTE is that most produce relatively severe focal or diffuse damage that is more similar to the type of injury found in moderate to severe rather than mild TBI. Significant efforts have been made to adapt these models to produce effects of mild TBI. For example, the CCI device has been modified to deliver impacts to the intact mouse skull resulting in a more mild TBI-like outcome [73]. The weight drop technique has also been used to produce repetitive mild TBI in the mouse [74,75], and the lateral fluid percussion model has been modified to deliver milder injuries in the rat [76]. Additional models have been developed to mimic mild TBI in combat conditions including a closed-head projectile concussive impact in rats [77], and a model of blast induced repetitive mild TBI in the rat that has been found to induce chronic behavioral changes [78,79].

Animal models of single-episode TBI have been valuable in determining the time course and the extent of cytoskeletal derangements which are a key feature of CTE. In both humans and experimental animals, TBI-induced axonal damage is associated with extensive accumulation of multiple proteins [65,81]. Axonal microtubule and

neurofilament (NF) proteins are altered acutely after TBI [82], and they are more pronounced following moderate injury [83]. In pigs subjected to a rotational acceleration injury, diffuse axonal damage is characterized by co-accumulation of tau and A β together with NF and APP throughout the white matter, while in cortical regions phospho-tau aggregates appear in axonal bulbs as well as in structures resembling NFT [81]. Similar changes occur in rodent models of TBI in which alterations in tau and other microtubule associated proteins can be rapid and transient [84,85]. For example, in rats subjected to a mechanical compression injury increased tau phosphorylation occurs in cortical regions in as little as 10 min following injury but then is undetectable by 12 hours [84]. Transient elevations of serum and CSF tau can also be detected following TBI in rats [86,87]. More sustained increases in a cleaved form of tau have been found in the cortex and hippocampus of rats following a CCI injury [85], and increased phospho-tau has been observed in cortex of mice 30 days after 5 mild TBI injuries were delivered utilizing a weight drop device [75]. A single blast-induced TBI resulted in acute (up to two weeks) axonal injury in the form of silver impregnated neuronal processes in rats [88] and increased levels of phosphorylated tau were detected in NFT like lesions in mice [58]. In rats, increased levels of tau have been found in multiple brain regions at 71 days following a blast injury [89]. Thus acute to subacute changes in tau are common across a spectrum of experimental TBI models.

However, few studies have yet revealed whether a subsequent chronic neurodegenerative changes are induced. Indeed the only study to our knowledge that has addressed this issue is the study by Hoshino *et al.* [90] who studied the chronic effects of fluid percussion injury in rats. They reported a progressive loss of cortical neurons over a six-month period accompanied by appearance of increasing numbers of phospho-tau immunoreactive neurons [90]. As noted above, rodent models of TBI are widely used because of their accessibility and relatively low cost. However, in addition to the anatomic differences between rodent and human brain, rodent models for CTE are potentially limited by the fact that mice and rats do not readily develop human like neurofibrillary pathology, a key histopathological feature of CTE. Rodents and humans differ in life spans, and species differences in tau also contribute to the difficulty of inducing NFT like lesions in rodents. The lack of human-like NFT pathology in rats and wild type mice has in particular been a limiting factor in the development of CTE models and has led to attempts to create more human like tauopathy models by introducing human tau transgenes into mice.

The non-traumatic neurodegenerative diseases that most closely resemble CTE pathologically are some forms of

frontotemporal dementia (FTD). While most FTD cases can occur without evidence of family history, in some families the disease is inherited and caused by mutations in the tau gene on chromosome 17 [91]. Mice expressing either human wild type or FTD-mutant tau develop a neurodegenerative disease spontaneously and accumulate NFT-like lesions with aging [92]. These lines have been used to investigate tauopathy-related dementias and offer potentially relevant models for CTE [92]. However to date only limited use has been made of these mice to study TBI. One study [93] examined the effects of mild repetitive TBI using a CCI injury in transgenic mice expressing the shortest human tau isoform. Transgenic and non-transgenic mice were subjected to a total of 16 mild TBI injuries over a period of 4 weeks and animals were examined histopathologically at 9 months after injury. Despite this aggressive injury protocol there was no general effect on histopathology or behavior of the tau transgene other than in one of the 18 transgenic mice that showed behavioral deficits and developed extensive telencephalic NFT like lesions and cerebral atrophy. In contrast, increases in total and phospho-tau immunoreactivity have been seen following a more severe CCI injury in JPNL mice which express the four repeat form of human tau containing the P301L mutation associated with FTD [94]. In another study, mice that expressed the 6 isoforms of human tau and lacked murine tau were subjected to a single or repetitive mild TBI using an electromagnetically controlled impactor; three weeks later, phospho-tau immunoreactivity, neuronal injury, and glial reaction were more pronounced in the group with repetitive mild TBI [95].

As discussed above, CTE is a heterogeneous disease with inconsistent pathology including not only tau but also A β lesions. Accordingly, several studies have investigated the effects of TBI in 3xTg-AD mice [94,96,97]. These mice also express the P301L mutation in tau associated with FTD but in combination with APP and presenilin mutations associated with familial Alzheimer's disease. With aging these mice develop increased A β 40 and A β 42 levels, accumulate intraneuronal A β and exhibit amyloid plaques and NFT like lesions, thus exhibiting a more complete spectrum of AD like pathology when compared to other transgenic mouse models. Controlled cortical impact injury in 3xTg-AD mice accelerated the development of tau abnormalities [96] with increased phospho-tau immunoreactivity seen histologically in the days following injury [96]. Tau changes persisted for at least 7 days after injury and were associated with intra-axonal accumulation of several kinases that phosphorylate tau [96]. In agreement with previous reports in other transgenic mouse models [98-100], the CCI injury in 3xTg-AD mice also resulted in intra-axonal A β accumulation [96]. However, in this model combining tauopathy and amyloidosis, the anatomic

pattern and time course of changes in A β and tau were distinct [96], and treatment with a γ -secretase inhibitor blocked posttraumatic A β accumulation but had no effect on tau pathology suggesting that once initiated these two pathologies may take independent courses of progression [96]. In contrast, reports from Loane *et al.* indicate that A β -reducing agents prevented TBI-induced neurodegeneration [101,102].

Better animal models of CTE are an urgent need given that to date no injury in animals has replicated the full spectrum of pathological findings in human CTE. Acute to subacute changes in tau are common across a range of experimental TBI models suggesting that injured animals can manifest at least some initial cytoskeletal changes observed in brains of humans with CTE. However most studies have focused on acute effects of TBI and greater attention should be directed to determining whether a chronic neurodegenerative condition can be induced after TBI in animals. As mild TBI and concussions draw increasing attention as the most common type of civilian and military TBI, future studies of repetitive concussions in both rodent and non-rodent species will be needed to determine the optimal model.

Neuroimaging of CTE

Identification of specific imaging biomarkers for CTE has been challenged by the lack of correlation between any available neuroimaging studies and post-mortem tissue assessments demonstrating histopathologic hallmarks of CTE. To date, no systemic *in vivo* biological markers have been identified that reveal the presence of CTE. Given that most case reports of CTE involve individuals with a history of repetitive sports-related trauma, the neuroimaging portion of this review will focus upon abnormalities present in athletes with a history of concussion.

Common structural abnormalities often reported in the brain of professional boxers include cavum septum pellucidum (often with fenestrations), ventricular and sulcal enlargement, and cortical and cerebellar atrophy. Although CSP is often reported in studies of boxers and is postulated to be a sign of DP/CTE [43], and a marker of brain atrophy [44,103,104], its relatively high incidence in the normal adult population precludes its presence as a diagnostic specific to boxing [105]. Despite the lack of specific *in vivo* findings in CTE, studies using both traditional and newer imaging methods have advanced our understanding of the consequences of repetitive head trauma. Newer imaging modalities and the use or development of advanced analytical methods (e.g., ref [106]) have revealed brain abnormalities in repetitive head trauma where more traditional methods found none, suggesting that comprehensive evaluation of CTE should include neuroimaging as part of an overall clinical assessment.

Structural Imaging

The major traditional structural imaging techniques used to evaluate brain function in boxers, and more recently in football players, are computed tomography (CT) and magnetic resonance imaging (MRI). In acute settings, these standard, structural imaging techniques have clinical utility in ruling out other acquired brain disorders such as tumor or stroke and have been utilized to rule out potentially fatal consequences of acute TBI such as hemorrhagic contusion, subdural and epidural hematomas, and subarachnoid hemorrhage. However, structural (or functional for that matter) imaging findings alone are not sufficient to diagnose CTE or determine who might eventually develop CTE. In addition, some of the structural imaging changes identified in CTE are similar to those seen in AD [107], and there may be similarities as well with normal aging. Early studies demonstrated that CT might not be as effective as MRI in identifying injury in the acute phase [108]. In non-acute settings, high-field MRI may be the preferred modality in the evaluation of TBI due to its improved tissue clarity and contrast of pathology, and because it eliminates the risk of ionizing radiation associated with CT [103,107,109]. These features of MRI make it a valuable diagnostic tool for determining the extent of chronic TBI [107].

In view of the improved tissue contrast and additional biologically relevant information afforded by MRI, early studies began to investigate whether MRI was superior to CT as an imaging modality in boxers. A study conducted in 1990 did not demonstrate MRI superiority to CT in 47 retired amateur boxers as compared to two age-matched control groups: soccer and track and field athletes [110]. However, another study published that same year found that some abnormalities detected by MRI were not shown by CT [103]. There were several limitations in these early studies, including comparison of professional boxers to amateur boxers, who may not show neurologic damage because of their limited number of exposures, and incomplete information in professional boxers in terms of number of years spent boxing or the number of bouts fought. In addition, most studies were cross-sectional, thus it cannot be determined from these studies whether the abnormalities were present before or the result of, boxing [105]. It should be noted that both MRI and CT have seen dramatic technological improvements over the past two decades.

A recent review of the MRI literature [107] used a systematic checklist approach to assess 100 unselected consecutive 1.5 and 3.0 Tesla MRI examinations of professional boxers to determine the extent of identifiable TBI findings. Results indicated that 76% of boxers had at least one finding associated with TBI: 59% had hippocampal atrophy, 43% CSP, 32% dilated perivascular spaces, 29% diffuse axonal injury, 24% cerebral atrophy,

19% increased lateral ventricular size, 14% pituitary gland atrophy, 5% arachnoid cysts, and 2% had contusions [107] (Figures 7 and 8). Statistical relationships were found between number of bouts and lateral ventricular size, and years of fighting correlated with dilated perivascular spaces and diffuse axonal injury. An earlier systematic review of the literature from 1985 to 2000 [111] on concussion in a variety of contact sports, found that male boxers had the highest frequency of concussion at the recreational level [111]. While these reviews indicate significant brain abnormalities in boxers exposed to frequent concussive and subconcussive blows to the head, prospective studies will be needed to determine whether the underlying process is degenerative and progressive in nature and related to and consistent with CTE clinical outcomes.

Diffusion tensor imaging

Newer imaging techniques such as diffusion tensor imaging (DTI) (Figure 9) are more sensitive for identifying the presence of axonal injury that occur due to the shearing forces of TBI. DTI is a relatively new technique and provides information reflecting the integrity of white matter fiber tracts.

Quantitative values used for spatially mapping DTI include apparent diffusion coefficient (typically mean diffusivity, MD), which measures the magnitude of the diffusion of water molecules, and fractional anisotropy (FA) or relative anisotropy (RA), which measure the directional preference of water molecules throughout brain fibers. White matter tracts normally constrain the isotropic diffusion of water. An FA value approaching 1.0 reflects maximal anisotropic diffusion, and values approaching zero indicate compromised white matter integrity. In addition to FA and RA, axial diffusivity and

radial diffusivity describing magnitude of diffusion longitudinal and transverse to the dominant direction of white matter tracts may also be derived from the calculated diffusion tensor. DTI values are affected when there are changes to the microstructure of brain tissue. Disruption of axonal integrity due to shearing forces associated with head trauma results in a general reduction of FA values as the normal barrier to isotropic diffusion is altered. This type of information is not available from routine MRI; thus, white matter abnormalities that may be present due to repetitive TBI often go undetected with routine imaging.

Chappell *et al.* [113] used a voxel-based analysis of DTI data to examine the ADC and FA in professional boxers and found previously unreported abnormalities that were assumed to reflect cumulative brain injury resulting from non-severe, repetitive head trauma [113]. Regions were found to have increased ADC, decreased FA, and decreased ADC in gray matter in the boxer group compared to a group of controls with no neurological history. The regions affected included lower splenium and cortical regions laterally and dorsolaterally in the frontal and posterior lobes. The authors concluded that their findings represented evidence that sustained boxing activity causes structural abnormalities in the brain.

Similarly, an earlier study by Zhang and colleagues [114] using quantitative diffusion weighted imaging (DWI) found increases in the whole brain diffusion constant in professional boxers as compared to age-matched, non-boxing control subjects. The increased diffusion values were observed despite negative or nonspecific results on routine MR imaging, indicating that this imaging modality may show early pathologic changes in the cellular and microvascular structure of the brain in boxers. Zhang *et al.* [114] postulated that given the similar findings in

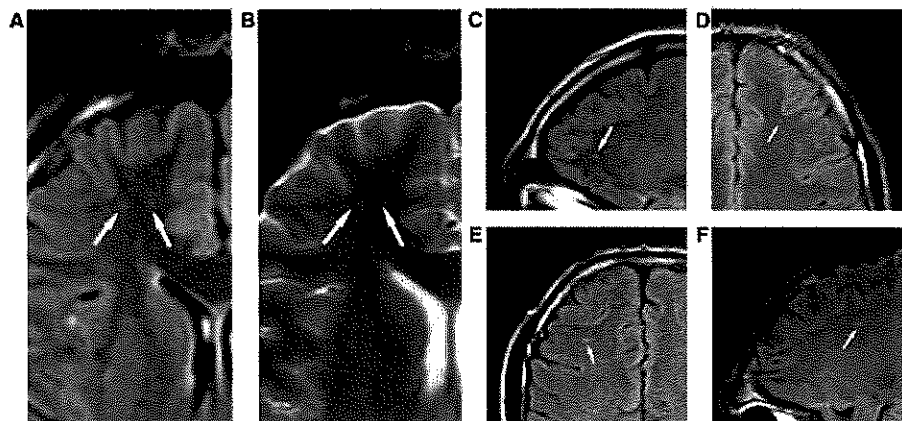
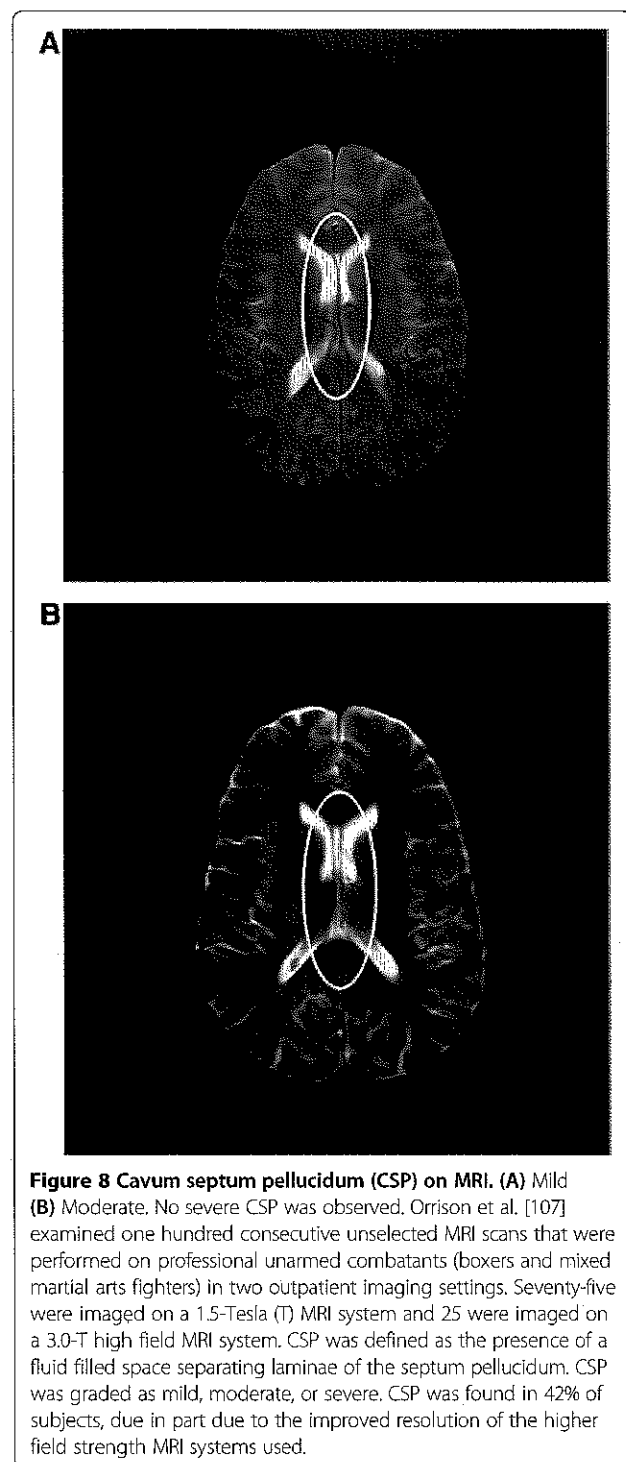
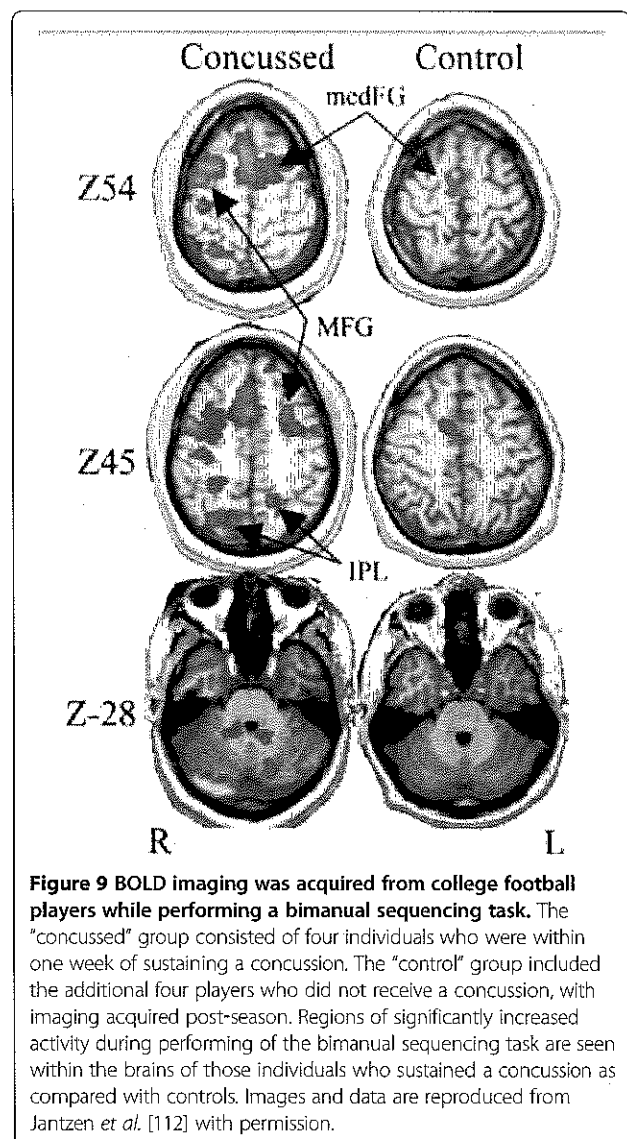


Figure 7 Diffuse axonal injury on MRI. (Panels A-F) Typical diffuse axonal injury; indicated by arrow. DAI was defined as focal areas of abnormal increased signal intensity on FLAIR and T2-weighted sequences, measuring up to 5 mm in maximum diameter, and located at the gray matter=white matter interface or within or adjacent to the corpus callosum. In the Orrison *et al.* sample, 29% had DAI. Reproduced from Orrison *et al.* [107] with permission.



patients with dementia, their results suggested that the whole brain diffusion coefficient represents preclinical signs of cognitive decline [114]. A later study by Zhang *et al.* [115] using DTI in a sample of 49 professional boxers and 19 healthy controls focused on the corpus callosum and the internal capsule. In that study, 42 boxers (86%) had a normal routine, clinical MRI and 7



had nonspecific white matter disease. In the boxer group, the whole brain diffusion constant was increased and FA was decreased significantly in the CC and posterior limb of IC, which the authors postulate may represent preclinical signs of subtle brain injury in professional boxers. Importantly, the increases in diffusion were found without brain abnormalities revealed on standard MR images, suggesting that diffusion tensor imaging is an important tool in the monitoring of brain function in professional boxers [113-115] and may be particularly useful in the multivariate diagnostic evaluation of individuals with CTE (Figure 10).

Functional magnetic resonance imaging (fMRI)

fMRI involves acquisition of a blood oxygen level dependent (BOLD) sequence during performance of a specific task (Figure 11). The BOLD sequence is sensitive to

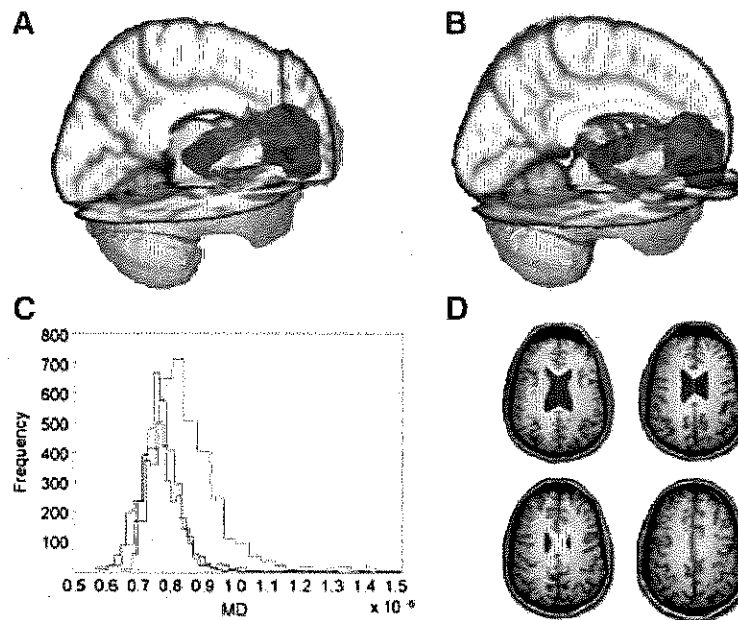


Figure 10 Diffusion tensor imaging (DTI) of TBI. Diffusion weighted volumes in 64 directions were collected from 22 patients with TBI (18 moderate/severe and 4 mild based upon Mayo classification system for TBI severity) and from 21 age-matched controls. Processing for DTI was performed and anatomic regions-of-interest were identified in controls for performing probabilistic tractography to study thalamo-cortical connections. Templates were generated that included white matter tracts isolated from probabilistic tractography. All brains were transformed into standard space and templates were applied to TBI patients and healthy controls to explore differences in white matter integrity between groups. Patients with a small (A) and large (B) number of abnormal voxels are shown. Histograms of MD values (C) from the patient shown in frame A (blue), frame B (red), as compared with a mean atlas (D) are shown. Images and data are reproduced from Squarcina et al. [116] with permission.

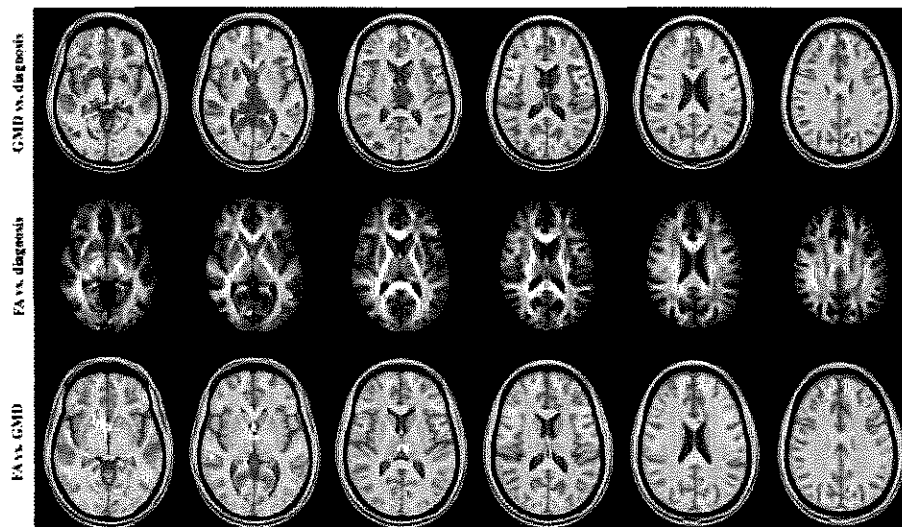


Figure 11 Sparse canonical correlation analysis (SCCA) of T1-weighted MP-RAGE and 30-direction diffusion tensor images (DTI) datasets are used to quantify traumatically induced disruption of WM and cortical networks. The cohort includes 17 controls and 16 patients with TBI (age and gender matched). Each patient had a history of non-penetrating TBI of at least moderate severity. White matter integrity is assessed by DTI and fractional anisotropy (FA) maps are generated. Separately, probabilistic segmentation of the T1-weighted imaging is performed to assess gray matter integrity. Variation in brain shape across subjects is normalized by diffeomorphically mapping these data into a population-specific template space. Image processing steps rely on the Camino and ANTs (Advanced Normalization Tools) neuroimage analysis open source toolkits. SCCA demonstrates significant differences between the control and patient groups in both the FA ($p < 0.002$) and gray matter ($p < 0.01$) that are widespread but largely focus on thalamocortical networks related to the limbic system. Using SCCA identified regions, a strong correlation is identified between degree of injury in WM and GM within the patient group. Figure courtesy of James R. Stone.

conformational changes in hemoglobin and can demonstrate regional variations in blood oxygenation. It is believed these changes in blood oxygenation reflect variations in neuronal activity. However, this point is a subject of controversy given the potential for the neurovascular apparatus to be altered in states of disease.

Distinct regional differences in BOLD signal have been reported in multiple studies of concussed athletes vs. controls. Chen *et al.* [117] utilized fMRI with a working memory task to study 16 athletes (13 hockey players, 2 wrestlers, one snowboarder), most of whom had a history of > 5 career concussions. An interval of between 1 and 14 months had passed between the scan and time of last concussion. Although no differences in task performance were seen between groups, significantly different regional patterns of activation were seen within the mid-dorsolateral prefrontal cortex (MDPC) in the concussed athletes as compared to controls. In a separate study, Chen *et al.* [118] utilized an fMRI working memory task to demonstrate abnormal activation patterns in the MDPC in 9 concussed athletes as compared to controls. Of note, no significant differences were seen with routine structural imaging or behavioral outcomes between groups. Lovell *et al.* [119] employed an N-back working memory task to evaluate 28 athletes within a mean of 6.6 days after sustaining a concussion. A components analysis identified three distinct networks that were activated in both concussed athletes and controls during participation in the N-back task. One of these networks involving Brodman's area 6 demonstrated increased levels of activity in concussed athletes compared with controls. Of note, the degree of increased activation in Brodmann's area 6 in the concussed athletes correlated significantly with prolonged recovery time and delayed return to play.

Positron emission tomography (PET)

PET imaging allows for characterization of the uptake and spatial localization of radiotracers injected intravenously (IV). This imaging modality allows for the sensitive detection of small molar concentrations of radiotracers given its sensitivity for detecting the high-energy photons (512 keV) produced by radioactive decay through pair production. PET imaging in sports related TBI is currently limited and has generally been confined to use of ^{18}F fluoro-deoxyglucose (^{18}F FDG), a glucose analog that reflects metabolism. In brain, where glucose is the sole energy source, ^{18}F FDG is an excellent tracer for this purpose. Provenzano *et al.* [120] employed ^{18}F FDG PET to study 19 boxers, aged 20 to 38 yrs, and compared them to 7 age-equivalent controls. The boxers had participated in an average of 17.3 matches, had performed poorly in a match or had been knocked out, and were referred for assessment because of clinical signs of neurological impairment. The PET scans were acquired as an element of the

clinical work-up. Retrospective evaluation of these scans was performed. Specific regions of hypometabolism were seen in the boxers' brains as compared to those of the controls, including the posterior cingulate cortex, parieto-occipital cortex, frontal lobes, and cerebellum.

In addition to assessing regional metabolism, PET can be used to detect the presence of specific molecules or abnormal cellular processes (molecular imaging). A β -deposition is an inconsistent feature in CTE. Diffuse plaques are found in about half (47%) of neuropathologically verified CTE cases [21], a figure that represents a greater frequency than that observed acutely after severe TBI (~30%) [51]. PET imaging using A β probes are being currently used in large scale studies of AD. The use of amyloid imaging in suspected CTE has not been established but will be of interest given the similarity between AD and CTE phenotypes and may be useful in determining whether the presence of amyloid post TBI can help predict outcome. This will be of particular interest if the presence of amyloid is associated with worse outcomes, since interventions that reduce the trajectory of brain amyloid accumulation have already been identified in studies of AD.

The original Corsellis series identified diffuse amyloid in 50% of the cases of DP. As mentioned above, DeKosky reported acute amyloidosis within hours or days even in young patients [51]. The natural history of these deposits remains to be established, and this is an important question that can now be addressed with amyloid imaging. Menon has recently reported just such a study, and his conclusion is that the initial burst of amyloidosis is rapidly cleared [121]. One wonders, however, whether clearance might become less efficient as repeated TBI occurs, and longer term studies of such a possibility are underway in our center. In another study, about half of subjects in the chronic phase of recovery from a severe TBI showed positive amyloid scans [122].

Of particular relevance to CTE, PET imaging ligands are now in development for detection of abnormally phosphorylated tau associated with neurofibrillary tangles (Figure 5). Zhang *et al.* [19] recently reported development of an ^{18}F labeled compound (^{18}F T807) which demonstrated high affinity and selectivity for tau in competitive assays designed to assess tau binding in post-mortem human brain slices derived from individuals with AD. This study also reported high uptake and washout in rodent brains, suggesting adequate blood brain barrier penetration. Given the presence of abnormally phosphorylated tau tangles in CTE, there is great interest in exploring the utility of this probe as a potential *in vivo* marker for CTE. Successful development of a molecular imaging ligand for neurofibrillary tangles would open the door for epidemiological studies to assess the true prevalence of this disease, correlative studies to determine other potential imaging diagnostics for CTE, and as a

potential metric to determine the efficacy of therapeutic or mitigation strategies for the treatment or prevention of CTE. Recent reports indicate that [^{18}F] T807 has been successfully employed for the detection of CTE tauopathy *in vitro* and *in vivo* [123-125] (Figure 12).

In summary, neuroimaging in CTE holds promise in serving as an *in vivo* biomarker and should be used in adjunct with a comprehensive history and thorough clinical diagnostic evaluation in life. Prospective, longitudinal neuroimaging studies are needed to help define the clinical and biological markers associated with, and

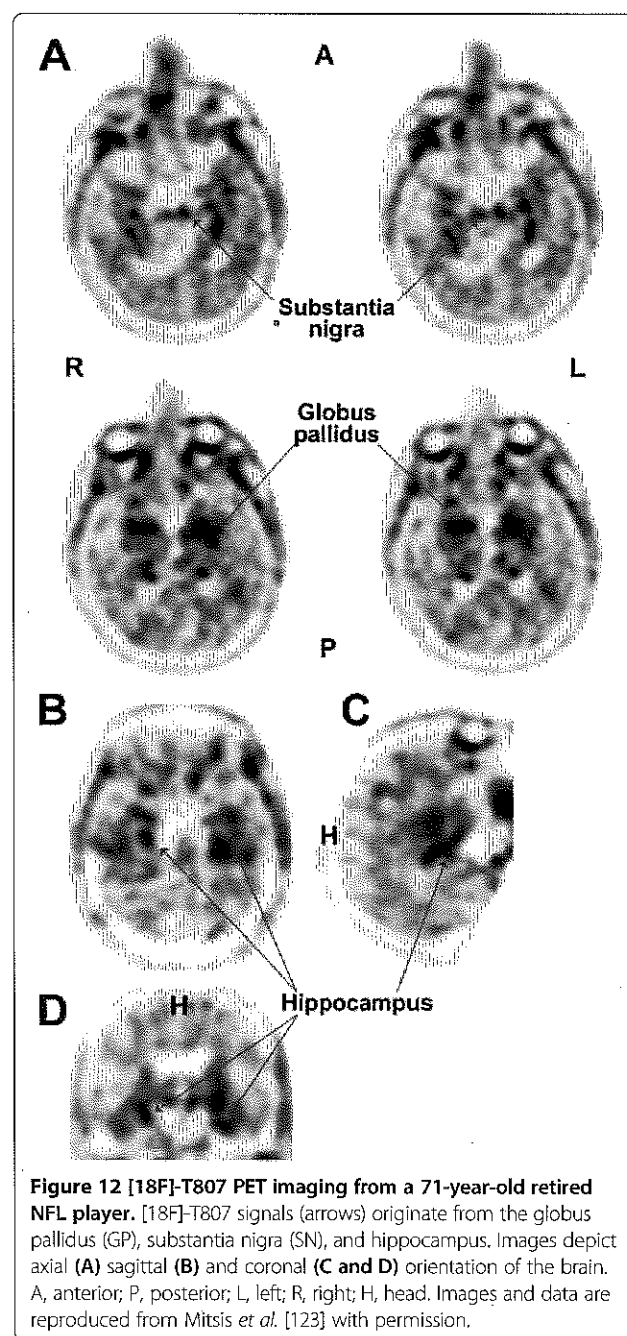
specific to, CTE. Body fluid markers may eventually complement neuroimaging markers, especially in medical centers without access to PET. These body fluid markers have recently been reviewed in detail elsewhere [126].

Conclusions

The association of CTE with certain sports and with battlefield blast exposure provides a mandate for the construction of accurate databases and epidemiological studies. Securing valid incidence, prevalence, and relative risk data are essential. Current academic papers continue to meet with criticism that the risk is being overstated [127,128]. While awaiting accurate data upon which to estimate relative risk, informed consent should be developed and offered to those contemplating exposure to repetitive TBI. While the administrative organizations of high impact sports and the military are the obvious organizations to begin informing potential athletes and recruits about the possible risk of CTE, another significant challenge will be the development of informed consent procedures for those overseeing the exposure of children and adolescents [27]. Finding the right balance of established fact and informed opinion will be a challenge, so as not to create unnecessary concern. Extension of genetic epidemiology studies to the pediatric and adolescent population will also be a challenge, albeit a worthy one [27]. The acquisition of definitive data about what genetic factors identified in adolescents are predictive of the eventual personal risk of CTE will require decades of study but are essential.

In parallel, application of validated laboratory models and execution of human clinical trials in the TBI/CTE area should be accelerated. Adaptation of repetitive TBI systems to the mouse holds promise for elucidating the molecular pathogenesis of CTE. The role of APOE genotype should be given high priority in both the animal and the human clinical studies [25,27]. Compounds emerging from drug discovery efforts in academic and pharmaceutical neurodegenerative disease programs should be considered for assessment in TBI [100]. Emerging data from the AD literature indicating the key role of immune-inflammatory processes may illuminate CTE as well. New data implicate IL-1 β in facilitating the development of tauopathy while reducing amyloidosis [68]. Tau oligomerization [129] may also contribute to the pathogenesis of CTE and may emerge acutely or after a delay [90]. Taken together with the A β independence of progressive tauopathy as reported by Brody [96], the hypothesis that tauopathy and amyloidosis can be dissociated in TBI is supported. The strong association of PTSD with TBI should also be evaluated and clarified [79].

The challenge of accelerating molecular neuropathology research in an era of dwindling resources is a major concern. Recent alliances between the NFL and the NIH,



between the NFL and General Electric, and between the Department of Defense and the NIH Alzheimer's Disease Neuroimaging Initiative provide some optimism that all the stakeholders are becoming engaged. Permanent commitment and substantial resources from these stakeholders will be required to offset the general reductions in federal support (including NIH and the Department of Defense) due to policies resulting in the recent sequestration of federal funds. In the absence of reliable prevalence data, the calculation of the potential healthcare savings by reduction of TBI exposure is not possible. The total cost of TBI exposure, of course, must take into account what may be enormous payouts from potential class action litigation verdicts. In addition to significant fiscal savings, the support of translational and clinical research programs in TBI/CTE and of development of policies that mitigate some or all of the attributable dementia risk represented by TBI/CTE is obviously justifiable on compassionate grounds. TBI, whether deliberate or unavoidable exposures, represents a source of pain and suffering that could be eliminated or at least greatly reduced with the pooled commitment and resources of the medical, sports, and military sectors. The high visibility of CTE in the media in the early 21st century makes this an opportune moment to apply the information reviewed herein so as to make important advances in elucidation of pathogenesis and in drug discovery.

Competing interests

The authors declare that they have no competing interests.

Authors' contributions

SG and STD summarized the history of CTE, wrote the overview and the neuropathology and PET imaging sections, and they were responsible for the overall organization, collation and final editing of the paper. GE, MDI, and STA drafted the experimental pathology sections. EM and JRS drafted the MRI sections. JB and EM drafted the neuropsychology section. All authors read and approved the final manuscript.

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References

- Martland HS: Punch drunk. *JAMA* 1928, **91**:1103-1107. doi:10.1001/jama.1928.02700150029009.
- Millsbaugh JA: Dementia pugilistica. *US Naval Med Bull* 1937, **35**:297-303.
- Critchley M: Medical aspects of boxing, particularly from neurological standpoint. *Brit Med J* 1957, **1**:357-362.
- Corsellis JAN, Bruton CJ, Freeman-Browne D: The aftermath of boxing. *Psychol Med* 1973, **3**:270-303.
- Saing T, Dick M, Nelson PT, Kim RC, Cribbs DH, Head E: Frontal cortex neuropathology in dementia pugilistica. *J Neurotrauma* 2012, **29**(6):1054-1070. doi:10.1089/neu.2011.1957.
- Brain Injury in Boxing: Council on Scientific Affairs. *JAMA* 1983, **249**(2):254-257. doi:10.1001/jama.1983.03330260072039.
- Boxing participation by children and adolescents. *Pediatrics* 2011, **128**:617-623. doi:10.1542/peds.2011-1165.
- N.Y. MMA legalization push continues. [http://espn.go.com/new-york/story/_/id/8881747/association-boxing-commissions-working-legalize-mma-new-york]
- Dementia Pugilistica. http://en.wikipedia.org/wiki/Dementia_pugilistica. At this URL, Wikipedia maintains a semi-comprehensive list of famous boxers, American football players, hockey players, and wrestlers believed to have been affected by DP/CTE. The following summary of famous persons with DP/CTE is reprinted with permission: Boxing: Rocky Graziano, Jimmy Ellis, Floyd Patterson, Bobby Chacon, Jerry Quarry, Mike Quarry, Jimmy Young, Wilfred Benitez, Emile Griffith, Willie Pep, Freddie Roach, Sugar Ray Robinson, Billy Conn, Joe Frazier, Fritzie Zivic, Meldrick Taylor, Muhammad Ali. American football: As of December 2012, thirty-three former National Football League (NFL) players have been diagnosed post-mortem with CTE, among them Mike Webster, Terry Long, Andre Waters, Justin Strzelczyk, Tom McHale, Former Detroit Lions lineman, Pro Bowler Lou Creekmur, former Houston Oilers/Miami Dolphins linebacker John Grimsley, former Tampa Bay Buccaneers guard Tom McHale, former Cincinnati Bengals wide receiver Chris Henry, former Chicago Bears safety Dave Duerson, former Buffalo Bills star running back Cookie Gilchrist, Wally Hilgenberg, Colt tight end John Mackey, former San Diego Charger Junior Seau, and former Atlanta Falcons safety Ray Easterling. Ice hockey: Reg Fleming, Rick Martin, Derek Boogaard. Wrestling: Chris Benoit, Andrew Martin.
- Omali BI, DeKosky ST, Minster RL, Kamboh MI, Hamilton RL, Wecht CH: Chronic traumatic encephalopathy in a national football league player. *Neurosurgery* 2005, **57**:128-134.
- NFL Picks New Chairmen for Panel on Concussions. [http://www.nytimes.com/2010/03/17/sports/17concussions.html?_r=0]
- Soccer Study Ties "Heading" to Brain Damage. [http://www.cbsnews.com/8301-504763_162-57332833-10391704/soccer-study-ties-heading-to-brain-damage/]
- In Debate About Fighting in Hockey, Medical Experts Weigh In. [http://www.nytimes.com/2011/12/13/sports/hockey/in-debate-about-fighting-in-hockey-medical-experts-weigh-in.html?pagewanted=all]
- Brain Ailments in Veterans Likened to Those in Athletes. [http://www.nytimes.com/2012/05/17/us/brain-disease-is-found-in-veterans-exposed-to-bombs.html?pagewanted=all]
- Omali B, Hammers JL, Bailes J, Hamilton RL, Kamboh MI, Webster G, Fitzsimmons RP: Chronic traumatic encephalopathy in an Iraqi war veteran with posttraumatic stress disorder who committed suicide. *Neurosurg Focus* 2011, **31**(5):E3.

16. Suicide in the NFL. [<http://www.psychologytoday.com/blog/two-takes-depression/201205/suicide-in-the-nfl>]
17. Has Athletes' Use of Performance Enhancing Drugs Played a Role in Suicides? [http://articles.philly.com/2012-05-13/sports/31690175_1_peds-concussions-study-of-retired-athletes]
18. Veterans and Brain Disease. [<http://www.nytimes.com/2012/04/26/opinion/kristof-veterans-and-brain-disease.html>]
19. Xia CF, Arteaga J, Chen G, Gangadhamath U, Gomez LF, Kasi D, Lam C, Liang Q, Liu C, Mocharla VP, Mu F, Sinha A, Su H, Szardenings AK, Walsh JC, Wang E, Yu C, Zhang W, Zhao T, Kolb HC: [18F]T807, a novel tau positron emission tomography imaging agent for Alzheimer's disease. *Alzheimers Dement* 2013, **9**(6):666–76.
20. Gavett BE, Cantu RC, Shenton M, Lin AP, Nowinski CJ, McKee AC, Stern RA: Clinical appraisal of chronic traumatic encephalopathy: current perspectives and future directions. *Curr Opin Neurol* 2011, **24**:525–531.
21. McKee AC, Stern RA, Nowinski CJ, Stein TD, Alvarez VE, Daneshvar DH, Lee HS, Wojtowicz SM, Hall G, Baugh CM, Riley DO, Kubilus CA, Cormier KA, Jacobs MA, Martin BR, Abraham CR, Ikezu T, Reichard RR, Wolozin BL, Budson AE, Goldstein LE, Kowall NW, Cantu RC: The spectrum of disease in chronic traumatic encephalopathy. *Brain* 2013, **136**(Pt 1):43–64. doi:10.1093/brain/aww307.
22. Omalu B, Bailes J, Hamilton RL, Kamboh MI, Hammers J, Case M, Fitzsimmons R: Emerging histomorphologic phenotypes of chronic traumatic encephalopathy in American athletes. *Neurosurgery* 2011, **69**:173–183.
23. Stern RA, Riley DO, Daneshvar DH, Nowinski CJ, Cantu RC, McKee AC: Long-term consequences of repetitive brain trauma: chronic traumatic encephalopathy. *PM R* 2011, **3**(Suppl 2):S460–S467.
24. Baugh CM, Stamm JM, Riley DO, Gavett BE, Shenton ME, Lin A, Nowinski CJ, Cantu RC, McKee AC, Stern RA: Chronic traumatic encephalopathy: neurodegeneration following repetitive concussive and subconcussive brain trauma. *Brain Imaging Behav* 2012, **6**:244–254.
25. Jordan BD, Relkin NR, Ravdin LD, Jacobs AR, Bennet A, Gandy S: Apolipoprotein E epsilon 4 associated with chronic traumatic brain injury in boxing. *JAMA* 1997, **278**:136–140.
26. DeKosky ST, Ikonomic MD, Gandy S: Traumatic brain injury—football, warfare, and long-term effects. *N Engl J Med* 2010, **363**(14):1293–1296.
27. Gandy S, DeKosky ST: APOE epsilon4 status and traumatic brain injury on the gridiron or the battlefield. *Sci Transl Med* 2012, **4**(134):134ed4.
28. Gavett BE, Stern RA, McKee AC: Chronic traumatic encephalopathy: a potential late effect of sport-related concussive and subconcussive head trauma. *Clin Sports Med* 2011, **30**:179–188. xi.
29. Barth J, Freeman JR, Broshek DK, Varney RN: Acceleration-deceleration sport-related concussion: the gravity of it all. *J Athl Train* 2001, **36**:253–256.
30. Hart J, Kraut MA, Wornack KB, Strain J, Didehban N, Bartz E, Conover H, Mansinghani S, Hanzhang L, Cullum CM: Neuroimaging of cognitive dysfunction and depression in aging retired national football league players: a cross sectional study. *JAMA Neurol* 2013, **70**(3):326–335.
31. Parker H: Traumatic encephalopathy ('punch drunk') of professional pugilists. *J Neurol Psychopathol* 1934, **15**:20–28.
32. Corsellis JA: Boxing and the brain. *BMJ* 1989, **298**:105–109.
33. Dale GE, Leigh PN, Luthert P, Anderton BH, Roberts GW: Neurofibrillary tangles in dementia pugilistica are ubiquitinated. *J Neurol Neurosurg Psychiatry* 1991, **54**:116–118.
34. Allsop D, Haga S, Bruton C, Ishii T, Roberts GW: Neurofibrillary tangles in some cases of dementia pugilistica share antigens with amyloid beta-protein of Alzheimer's disease. *Am J Pathol* 1990, **136**:255–260.
35. Hof PR, Bouras C, Buée L, Delacourte A, Perl DP, Morrison JH: Differential distribution of neurofibrillary tangles in the cerebral cortex of dementia pugilistica and Alzheimer's disease cases. *Acta Neuropathol* 1992, **85**:23–30.
36. Roberts GW, Allsop D, Bruton C: The occult aftermath of boxing. *J Neurol Neurosurg Psychiatry* 1990, **53**:373–378.
37. Roberts GW: Immunocytochemistry of neurofibrillary tangles in dementia pugilistica and Alzheimer's disease: evidence for common genesis. *Lancet* 1988, **2**:1456–1458.
38. Tokuda T, Ikeda S, Yanagisawa N, Ihara Y, Glenner GG: Re-examination of ex-boxers' brains using immunohistochemistry with antibodies to amyloid beta-protein and tau protein. *Acta Neuropathol* 1991, **82**:280–285.
39. Nowak LA, Smith GG, Reyes PF: Dementia in a retired world boxing champion: case report and literature review. *Clin Neuropathol* 2009, **28**:275–280.
40. Geddes JF, Vowles GH, Robinson SF, Sutcliffe JC: Neurofibrillary tangles, but not Alzheimer-type pathology, in a young boxer. *Neuropathol Appl Neurobiol* 1996, **22**:12–16.
41. Jordan BD, Kanik AB, Horwich MS, Sweeney D, Relkin NR, Petito CK, Gandy S: Apolipoprotein E epsilon 4 and fatal cerebral amyloid angiopathy associated with dementia pugilistica. *Ann Neurol* 1995, **38**:698–699.
42. Mitsis E, Riggio S, D'Antonio E, Goldstein M, DeKosky ST, Naidich T, Dellman B, Machac J, Elder G, Sano M, Gandy S, Gordon W: Flortetapir scanning excludes Alzheimer's disease in a retired NFL player with delayed cognitive impairment. *Hum Amyloid Imag Conf Abstr* 2013, <http://www.worldeventsforum.com/hai/current-program-2013.html>.
43. Bogdanoff B, Natter HM: Incidence of cavum septum pellucidum in adults: a sign of boxer's encephalopathy. *Neurology* 1989, **39**:991–992.
44. McCrory P: Cavum septi pellucidum—a reason to ban boxing? *Br J Sports Med* 2002, **36**:157–161.
45. Aviv RI, Tomlinson G, Kendall B, Thakkar C, Valentine A: Cavum septi pellucidum in boxers. *Can Assoc Radiol J* 2010, **61**:29–32.
46. Guterman A, Smith RW: Neurological sequelae of boxing. *Sports Med* 1987, **4**:194–210.
47. Roberts AJ: *Brain Damage in Boxers* London: Pitman Medical Scientific Publications. 1969.
48. Ross RJ, Cole M, Thompson JS, Kim KH: Boxers—computed tomography, EEG, and neurological evaluation. *JAMA* 1983, **249**:211–213.
49. Viano DC, Casson IR, Pellman EJ, Bir CA, Zhang L, Sherman DC, Boitano MA: Concussion in professional football: comparison with boxing head impacts—part 10. *Neurosurgery* 2005, **57**:1154–1172.
50. Braak H, Braak E: Evolution of the neuropathology of Alzheimer's disease. *Acta Neurol Scand Suppl* 1996, **165**:3–12.
51. Ikonomic MD, Uryu K, Abrahamson EE, Ciallella JR, Trojanowski JQ, Lee VM, Clark RS, Marion DW, Wisniewski SR, DeKosky ST: Alzheimer's pathology in human temporal cortex surgically excised after severe brain injury. *Exp Neurol* 2004, **190**:192–203.
52. Omalu BI, DeKosky ST, Hamilton RL, Minster RL, Kamboh MI, Shakir AM, Wecht CH: Chronic traumatic encephalopathy in a national football league player: part II. *Neurosurgery* 2006, **59**:1086–1092.
53. Mac Donald CL, Johnson AM, Cooper D, Nelson EC, Werner NJ, Shimony JS, Snyder AZ, Raichle ME, Witherow JR, Fang R, Flaherty SF, Brody DL: Detection of blast-related traumatic brain injury in U.S. military personnel. *N Engl J Med* 2011, **364**:2091–2100.
54. Morey RA, Haswell CC, Selgrade ES, Massoglia D, Liu C, Weiner J, Marx CE, Cemak I, McCarthy G, MIRECC Work Group: Effects of chronic mild traumatic brain injury on white matter integrity in Iraq and Afghanistan war veterans. *Hum Brain Mapp*.
55. Magnuson J, Leonessa F, Ling GS: Neuropathology of explosive blast traumatic brain injury. *Curr Neurol Neurosci Rep* 2012, [Epub ahead of print].
56. Mayorga MA: The pathology of primary blast overpressure injury. *Toxicology* 1997, **121**:17–28.
57. Cernak I: The importance of systemic response in the pathobiology of blast-induced neurotrauma. *Front Neurol* 2010, **1**:151.
58. Goldstein LE, Fisher AM, Tagge CA, Zhang XL, Velisek L, Sullivan JA, Upreti C, Kracht JM, Ericsson M, Wojnarowicz MW, Goletiani CJ, Maglakelidze GM, Casey NA, Moncaster JA, Minaeva O, Moir RD, Nowinski CJ, Stern RA, Cantu RC, Gelling J, Blusztajn JK, Wolozin BL, Ikezu T, Stein TD, Budson AE, Kowall NW, Chargin D, Sharon A, Saman S, Hall GF, Moss WC, Cleveland RO, Tanzi RE, Stanton PK, McKee AC: Chronic traumatic encephalopathy in blast-exposed military veterans and a blast neurotrauma mouse model. *Sci Transl Med* 2012, **4**(134):134ra60.
59. Blennow K, Jonsson M, Andreassen N, Rosengren L, Wallin A, Hellström PA, Zetterberg H: No neurochemical evidence of brain injury after blast overpressure by repeated explosions or firing heavy weapons. *Acta Neurol Scand* 2011, **123**:245–251. doi:10.1111/j.1600-0404.2010.01408.x.
60. Shahim P, Tegner Y, Wilson DH, Randall J, Skillbäck T, Pazooki D, Kalberg B, Blennow K, Zetterberg H: Blood biomarkers for brain injury in concussed professional ice hockey players. *JAMA Neurol* 2014, doi:10.1001/jamaneurol.2014.367.
61. Abel EL: Football increases the risk for Lou Gehrig's disease, amyotrophic lateral sclerosis. *Percept Mot Skills* 2007, **104**:1251–1254.
62. Chiò A, Traynor BJ, Swingle R, Mitchell D, Hardiman O, Mora G, Beghi E, Logroscino G: EURALS consortium. Amyotrophic lateral sclerosis and soccer: a different epidemiological approach strengthen the previous findings. *J Neurol Sci* 2008, **269**:187–188.

63. Povlishock JT: Traumatocally induced axonal injury: pathogenesis and pathobiological implications. *Brain Pathol* 1992, **2**:1-12.
64. Smith DH, Chen XH, Iwata A, Graham DI: Amyloid beta accumulation in axons after traumatic brain injury in humans. *J Neurosurg* 2003, **98**:1072-1077.
65. Uryu K, Chen XH, Martinez D, Browne KD, Johnson VE, Graham DI, Lee VM, Trojanowski JQ, Smith DH: Multiple proteins implicated in neurodegenerative diseases accumulate in axons after brain trauma in humans. *Exp Neurol* 2007, **208**:185-192.
66. Graham DI, Gentleman SM, Nicoll JA, Royston MC, McKenzie JE, Roberts GW, Mrak RE, Griffin WS: Is there a genetic basis for the deposition of beta-amyloid after fatal head injury? *Cell Mol Neurobiol* 1999, **19**:19-30.
67. Johnson VE, Murray L, Raghupathi R, Stewart J, Nicoll JA, MacKinnon MA, McIntosh TK, Graham DI: No evidence for the presence of apolipoprotein epsilon 4, interleukin-1 alpha allele 2 and interleukin-1 beta allele 2 cause an increase in programmed cell death following traumatic brain injury in humans. *Clin Neuropathol* 2006, **25**:255-264.
68. Ghosh S, Wu MD, Shafit SS, Kyriakides S, LaFerla FM, Olschowska JA, O'Banion MK: Sustained interleukin-1 beta overexpression exacerbates tau pathology despite reduced amyloid burden in an Alzheimer's mouse model. *J Neurosci* 2013, **33**:5053-5064.
69. Cernak I: Animal models of head trauma. *NeuroRx* 2005, **2**:410-422.
70. Marklund N, Hillered L: Animal modelling of traumatic brain injury in preclinical drug development: where do we go from here? *Br J Pharmacol* 2011, **164**:1207-1229.
71. Elder GA, Mitsis EM, Ahlers ST, Cristian A: Blast-induced mild traumatic brain injury. *Psychiatr Clin North Am* 2010, **33**:757-781. doi:10.1016/j.psc.2010.08.001.
72. Pandey DK, Yadav SK, Mahesh R, Rajkumar R: Depression-like and anxiety-like behavioural aftermaths of impact accelerated traumatic brain injury in rats: a model of comorbid depression and anxiety? *Behav Brain Res* 2009, **205**:436-442. doi:10.1016/j.bbr.2009.07.027.
73. Laurer HL, Bareyre FM, Lee VM, Trojanowski JQ, Longhi L, Hoover R, Saatman KE, Raghupathi R, Hoshino S, Grady MS, McIntosh TK: Mild head injury increasing the brain's vulnerability to a second concussive impact. *J Neurosurg* 2001, **95**:859-870.
74. DeFord SM, Wilson MS, Rice AC, Clausen T, Rice LK, Barabnova A, Bullock R, Hamm RJ: Repeated mild brain injuries result in cognitive impairment in B6C3F1 mice. *J Neurotrauma* 2002, **19**:427-438.
75. Kane MJ, Angoa-Perez M, Briggs DI, Viano DC, Kreipke CW, Kuhn DM: A mouse model of human repetitive mild traumatic brain injury. *J Neurosci Methods* 2012, **203**:41-49.
76. Shultz SR, Bao F, Omana V, Chiu C, Brown A, Cain DP: Repeated mild lateral fluid percussion brain injury in the rat causes cumulative long-term behavioral impairments, neuroinflammation, and cortical loss in an animal model of repeated concussion. *J Neurotrauma* 2012, **29**:281-294. doi:10.1089/neu.2011.2123.
77. Chen Z, Leung LY, Mountney A, Liao Z, Yang W, Lu XC, Dave J, Deng-Bryant Y, Wei G, Schmid K, Shear DA, Tortella FC: A novel animal model of closed-head concussive-induced mild traumatic brain injury: development, implementation, and characterization. *J Neurotrauma* 2012, **29**:268-280.
78. Ahlers ST, Vasserman-Stokes E, Shaughnessy MC, Hall AA, Shear DA, Chavko M, McCarron RM, Stone JR: Assessment of the effects of acute and repeated exposure to blast overpressure in rodents: toward a greater understanding of blast and the potential ramifications for injury in humans exposed to blast. *Front Neurol* 2012, **3**:32. doi:10.3389/fneur.2012.00032.
79. Elder GA, Dorr NP, De Gasperi R, Gama Sosa MA, Shaughnessy MC, Maudlin-Jeronimo E, Hall AA, McCarron RM, Ahlers ST: Blast exposure induces post-traumatic stress disorder-related traits in a rat model of mild traumatic brain injury. *J Neurotrauma* 2012, **29**:2564-2575. doi:10.1089/neu.2012.2510.
80. Taber KH, Warden DL, Hurley RA: Blast-related traumatic brain injury: what is known? *J Neuropsychiatry Clin Neurosci* 2006, **18**(2):141-145C.
81. Smith DH, Chen XH, Noraka M, Trojanowski JQ, Lee VM, Saatman KE, Leoni MJ, Xu BN, Wolf JA, Meaney DF: Accumulation of amyloid beta and tau and the formation of neurofibrillary inclusions following diffuse brain injury in the pig. *J Neuropathol Exp Neurol* 1999, **58**:982-992.
82. Fitzpatrick MO, Dewar D, Teasdale GM, Graham DI: The neuronal cytoskeleton in acute brain injury. *Br J Neurosurg* 1998, **12**:313-317.
83. Saatman KE, Graham DI, McIntosh TK: The neuronal cytoskeleton is at risk after mild and moderate brain injury. *J Neurotrauma* 1998, **15**:1047-1058.
84. Chen LJ, Wang YJ, Tseng GF: Compression alters kinase and phosphatase activity and tau and MAP2 phosphorylation transiently while inducing the fast adaptive dendritic remodeling of underlying cortical neurons. *J Neurotrauma* 2010, **27**:1657-1669.
85. Gabbita SP, Scheff SW, Menard RM, Roberts K, Fugaccia I, Zemlan FP: Cleaved-tau: a biomarker of neuronal damage after traumatic brain injury. *J Neurotrauma* 2005, **22**:83-94.
86. Liliang PC, Liliang CL, Lu K, Wang K, Weng HC, Hsieh CH, Tsai YD, Chen HJ: Relationship between injury severity and serum tau protein levels in traumatic brain injured rats. *Resuscitation* 2010, **81**:1205-1208.
87. Siman R, McIntosh TK, Soltesz KM, Chen Z, Neumar RW, Roberts VL: Proteins released from degenerating neurons are surrogate markers for acute brain damage. *Neurobiol Dis* 2004, **16**:311-320.
88. Garman RH, Jenkins LW, Switzer RC 3rd, Bauman RA, Tong LC, Swauger PV, Parks SA, Ritzel DV, Dixon CE, Clark RS, Bayir H, Kagan V, Jackson EK, Kochanek PM: Blast exposure in rats with body shielding is characterized primarily by diffuse axonal injury. *J Neurotrauma* 2011, **28**:947-959.
89. Kovacs E, Gyorgy AB, Kwon SK, Wingo DL, Kamnakhsh A, Long JB, Kasper CE, Agoston DV: The effect of enriched environment on the outcome of traumatic brain injury: a behavioral, proteomics, and histological study. *Front Neurosci* 2011, **5**:42.
90. Hoshino S, Tamaoka A, Takahashi M, Kobayashi S, Furukawa T, Oaki Y, Mori O, Matsuno S, Shoji S, Inomata M, Teramoto A: Emergence of immunoreactivities for phosphorylated tau and amyloid-beta protein in chronic stage of fluid percussion injury in rat brain. *Neuroreport* 1998, **9**(8):1879-1883.
91. Premi E, Padovani A, Borroni B: Frontotemporal lobar degeneration. *Adv Exp Med Biol* 2012, **724**:114-127.
92. Gotz J, Ittner L: Animal models of Alzheimer's disease and frontotemporal dementia. *Nat Rev Neurosci* 2008, **9**:532-544.
93. Yoshiyama Y, Uryu K, Higuchi M, Longhi L, Hoover R, Fujimoto S, McIntosh T, Lee VM, Trojanowski JQ: Enhanced neurofibrillary tangle formation, cerebral atrophy, and cognitive deficits induced by repetitive mild brain injury in a transgenic tauopathy mouse model. *J Neurotrauma* 2005, **22**:1134-1141.
94. Tran HT, Sanchez L, Esparza TJ, Brody DL: Distinct temporal and anatomical distributions of amyloid-beta and tau abnormalities following controlled cortical impact in transgenic mice. *PLoS One* 2011, **6**:e25475. doi:10.1371/journal.pone.0025475.
95. Ojo JO, Mouzon B, Greenberg MB, Bachmeier C, Mullan M, Crawford F: Repetitive mild traumatic brain injury augments tau pathology and glial activation in aged hTau mice. *J Neuropathol Exp Neurol* 2013, **72**:137-151.
96. Tran HT, LaFerla FM, Holtzman DM, Brody DL: Controlled cortical impact traumatic brain injury in 3xTg-AD mice causes acute intra-axonal amyloid-beta accumulation and independently accelerates the development of tau abnormalities. *J Neurosci* 2011, **31**:9513-9525. doi:10.1523/JNEUROSCI.0858-11.2011.
97. Tran HT, Sanchez L, Brody DL: Inhibition of JNK by a peptide inhibitor reduces traumatic brain injury-induced tauopathy in transgenic mice. *J Neuropathol Exp Neurol* 2012, **71**:116-129. doi:10.1097/NEN.0b013e3182456aed.
98. Smith DH, Nakamura M, McIntosh TK, Wang J, Rodriguez A, Chen XH, Raghupathi R, Saatman KE, Clemens J, Schmidt ML, Lee VM, Trojanowski JQ: Brain trauma induces massive hippocampal neuron death linked to a surge in beta-amyloid levels in mice overexpressing mutant amyloid precursor protein. *Am J Pathol* 1998, **153**:1005-1010.
99. Abrahamson EE, Ikonovic MD, Ciallella JR, Hope CE, Paljug WR, Isanski BA, Flood DG, Clark RS, DeKosky ST: Caspase inhibition therapy abolishes brain trauma-induced increases in Abeta peptide: implications for clinical outcome. *Exp Neurol* 2006, **197**:437-450.
100. Abrahamson EE, Ikonovic MD, Dixon CE, DeKosky ST: Simvastatin therapy prevents brain trauma-induced increases in beta-amyloid peptide levels. *Ann Neurol* 2009, **66**:407-414.
101. Loane DJ, Pocivavsek A, Moussa CE, Thompson R, Matsuo Y, Faden AI, Rebeck GW, Burns MP: Amyloid precursor protein secretases as therapeutic targets for traumatic brain injury. *Nat Med* 2009, **15**:377-379. doi:10.1038/nm.1940.
102. Loane DJ, Washington PM, Vardanian L, Pocivavsek A, Hoe HS, Duff KE, Cernak I, Rebeck GW, Faden AI, Burns MP: Modulation of ABCA1 by an LXR agonist reduces beta-amyloid levels and improves outcome after traumatic brain injury. *J Neurotrauma* 2011, **28**:225-236. doi:10.1089/neu.2010.1595.
103. Jordan B, Zimmerman R: Computed tomography and magnetic resonance imaging comparisons in boxers. *JAMA* 1990, **263**:1670-1674.
104. Jordan BD, Jahre C, Hauser WA, Zimmerman RD, Zarelli M, Lipsitz EC, Johnson V, Warren RF, Tsairis P, Folk FS: CT of 338 active professional boxers. *Radiology* 1992, **185**:509-512.

105. McCrory P, Zazryn T, Cameron P: **The evidence for chronic traumatic encephalopathy in boxing.** *Sports Med* 2007, **37**:467-476.
106. Chappell MH, Brown JA, Dalrymple-Alford JC, Ulug AM, Watts R: **Multivariate analysis of diffusion tensor imaging data improves the detection of microstructural damage in young professional boxers.** *Magn Reson Imaging* 2008, **26**:1398-1405.
107. Orrison WW Jr, Hanson EH, Alamo T, Watson D, Sharma M, Perkins TG, Tandy RD: **Traumatic brain injury: a review and high-field mri findings in 100 unarmed combatants using a literature-based checklist approach.** *J Neurotrauma* 2009, **26**:689-701.
108. Gandy SE, Snow RB, Zimmerman RD, Deck MD: **Cranial nuclear magnetic resonance imaging in head trauma.** *Ann Neurol* 1984, **16**:254-257.
109. Jordan BD: **Chronic traumatic brain injury associated with boxing.** *Semin Neurol* 2000, **20**:179-185.
110. Haglund Y, Bergstrand G: **Does Swedish amateur boxing lead to chronic brain damage? 2. A retrospective study with CT and MRI.** *Acta Neurol Scand* 1990, **82**:297-302.
111. Koh JO, Cassidy JD, Watkinson EJ: **Incidence of concussion in contact sports: a systematic review of the evidence.** *Brain Inj* 2003, **17**(10):901-917.
112. Jantzen KJ, Anderson B, Steinberg FL, Kelso JA: **A prospective functional MR imaging study of mild traumatic brain injury in college football players.** *AJNR Am J Neuroradiol* 2004, **25**(5):738-745.
113. Chappell MH, Ulug AM, Zhang L, Heltger MH, Jordan BD, Zimmerman RD, Watts R: **Distribution of microstructural damage in the brains of professional boxers: a diffusion MRI study.** *J Magn Reson Imaging* 2006, **24**:537-542.
114. Zhang L, Ravdin LD, Relkin N, Zimmerman RD, Jordan B, Lathan WE, Ulug AM: **Increased diffusion in the brain of professional boxers: a preclinical sign of traumatic brain injury?** *AJNR Am J Neuroradiol* 2003, **24**:52-57.
115. Zhang L, Heier LA, Zimmerman RD, Jordan B, Ulug AM: **Diffusion anisotropy changes in the brains of professional boxers.** *AJNR Am J Neuroradiol* 2006, **27**:2000-2004.
116. Squarcina L, Bertoldo A, Ham TE, Heckemann R, Sharp DJ: **A robust method for investigating thalamic white matter tracts after traumatic brain injury.** *Neuroimage* 2012, **63**(2):779-788. doi:10.1016/j.neuroimage.2012.07.016.
117. Chen JK, Johnston KM, Frey S, Petrides M, Worsley K, Pito A: **Functional abnormalities in symptomatic concussed athletes: an fMRI study.** *Neuroimage* 2004, **22**:68-82.
118. Chen JK, Johnston KM, Petrides M, Pito A: **Recovery from mild head injury in sports: evidence from serial functional magnetic resonance imaging studies in male athletes.** *Clin J Sport Med* 2008, **18**:241-247.
119. Lovell MR, Pardini J, Welling J, Collins MW, Bakal J, Lazar N, Roush R, Eddy WF, Becker JT: **Functional brain abnormalities are related to clinical recovery and time to return-to-play in athletes.** *Neurosurgery* 2007, **61**:352-359. discussion 359-360.
120. Provenzano FA, Jordan B, Tikofsky RS, Saxena C, Van Heertum RL, Ichise M: **F-18 FDG PET imaging of chronic traumatic brain injury in boxers: a statistical parametric analysis.** *Nucl Med Commun* 2010, **31**:952-957.
121. Hong YT, Veenith T, Dewar D, Outtrim JG, Mani V, Williams C, Pimlott S, Hutchinson PJ, Tavares A, Canales R, Mathis CA, Klunk WE, Aigbirhio FI, Coles JP, Baron JC, Pickard JD, Fryer TD, Stewart W, Menon DK: **Amyloid imaging with carbon 11-labeled Pittsburgh compound B for traumatic brain injury.** *JAMA Neurol* 2014, **71**(1):23-31. doi:10.1001/jamaneurol.2013.4847. PubMed PMID: 24217171.
122. Kawai N, Kawanishi M, Kudomi N, Maeda Y, Yamamoto Y, Nishiyama Y, Tamiya T: **Detection of brain amyloid β deposition in patients with neuropsychological impairment after traumatic brain injury: PET evaluation using Pittsburgh Compound-B.** *Brain Inj* 2013, **27**(9):1026-1031.
123. Mitsis EM, Riggio S, Kostakoglu L, Dickstein DL, Machac J, Delman B, Goldstein M, Jennings D, D'Antonio E, Martin J, Naidich TP, Aloysi A, Fernandez C, Seibyl J, DeKosky ST, Elder GA, Marek K, Gordon W, Hof PR, Sano M, Gandy S: **Tauopathy PET and amyloid PET in the diagnosis of chronic traumatic encephalopathies: studies of a retired NFL player and of a man with FTD and a severe head injury.** *Trans Psychiatry* 4:e441. doi:10.1038/tp.2014.91.
124. Kolb HC, Attardo G, Conway K, Gomez F, Liang Q, Lin Y-G, Siderowf A, Skovronsky DM, Mintun MA: **Detection of PHF-Tau pathology with T557, T726 and [18 F] T807 in brain sections from Alzheimer's and non-Alzheimer's tauopathy patients.** In *Program and Abstract Book of the 8th Annual Human Amyloid Imaging Meeting: January 15-17 2014; Miami*. 2014:21.
125. Mintun MA: **PET Imaging of Human Tau Deposits with F18-T807.** In *In the 4th Annual Traumatic Brain Injury Conference: 16-17 April 2014; Crystal City, VA*.
126. DeKosky S, Blennow K, Ikonovic M, Gandy S: **Acute and chronic traumatic encephalopathies: pathogenesis and biomarkers.** *Nat Rev Neurol* advance online publication 26 March 2013; doi:10.1038/nrneurol.2013.36.
127. **NFL Doctor Says Disease is Overstated.** [http://www.nytimes.com/2013/03/28/sports/football/doctor-for-nfl-says-study-overstates-effects-of-cte.html?pagewanted=all&_r=0]
128. **Report: NFL Doctor Recommended Removing CTE from Safety Sheet.** [http://www.cbssports.com/nfl/blog/eye-on-football/2196494/report-nfl-doctor-recommended-removing-cte-from-safety-sheet]
129. Hawkins BE, Krishnamurthy S, Castillo-Carranza DL, Sengupta U, Prough DS, Jackson GR, Dewitt DS, Kaye R: **Rapid accumulation of endogenous tau oligomers in a rat model of traumatic brain injury.** *J Biol Chem* 2013, **288**(23):17042-17050. doi:10.1074/jbc.M113.472746.

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Acute and chronic traumatic encephalopathies: pathogenesis and biomarkers

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Abstract

Over the past decade, public awareness of the long-term pathological consequences of traumatic brain injury (TBI) has increased. Such awareness has been stimulated mainly by reports of progressive neurological dysfunction in athletes exposed to repetitive concussions in high-impact sports such as boxing and American football, and by the rising number of TBIs in war veterans who are now more likely to survive explosive blasts owing to improved treatment. Moreover, the entity of chronic traumatic encephalopathy (CTE)—which is marked by prominent neuropsychiatric features including dementia, parkinsonism, depression, agitation, psychosis, and aggression—has become increasingly recognized as a potential late outcome of repetitive TBI. Annually, about 1% of the population in developed countries experiences a clinically relevant TBI. The goal of this Review is to provide an overview of the latest understanding of CTE pathophysiology, and to delineate the key issues that are challenging clinical and research communities, such as accurate quantification of the risk of CTE, and development of reliable biomarkers for single-incident TBI and CTE.

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Introduction

Approximately 1% of the population of developed countries experiences traumatic brain injury (TBI) each year.¹ The pathological consequences of TBI² have received increasing media attention following reports of progressive neurological dysfunction in athletes who have been exposed to repetitive concussions in high-impact sports (initially boxing and, in the past 8 years, American football), and the increasing number of war veterans presenting with TBI caused by explosive blasts. Improved battlefield emergency care and, consequently, improved survival means that soldiers who might previously have died from polytrauma now survive and can go on to develop substantial cognitive deficits. This principle applies most clearly to veterans of the recent wars in Iraq and Afghanistan. The US Department of Defense terms TBI the 'signature injury' among veterans from these wars.³

In this article, we provide an overview of the acute and long-term pathophysiology of single-incident and repetitive TBI, focusing in particular on chronic traumatic encephalopathy (CTE). We briefly discuss imaging as well as plasma and cerebrospinal fluid (CSF) biomarkers for this disorder, and finally consider preventive measures that can be undertaken to avoid such injury. Understanding of the issues discussed will assist clinical and research communities to manage the risk of CTE and avoid disease.

Single-incident TBI

Acute sequelae

Many complications of TBI are evident immediately or soon after the injury.⁴⁻¹⁴ Acute post-traumatic sensory, motor and neurocognitive syndromes often occur owing to brain contusions, intracerebral haemorrhage, and axonal shearing. Skull fracture further complicates the pathobiology, leading to direct brain shearing and haemorrhage if the fracture is severe or depressed, and increasing the risk of ischaemia from vasoconstriction precipitated by blood products, seizures and infection. Seemingly mild 'closed-head' TBI, in which the skull is not fractured, can lead to diverse and sometimes disabling symptoms such as chronic headaches, dizziness and vertigo, difficulty in concentrating, word-finding problems, depression, irritability and impulsiveness. The duration of such symptoms is variable, but can be months or longer.

Post-traumatic stress disorder (PTSD) is a frequent accompaniment of military-related TBI, especially severe cases.⁵⁻¹⁴ Each syndrome can occur without the other: PTSD can occur following events of severe stress, and TBI can occur, even in combat, without resulting in PTSD. Their relationship, and how to differentiate and treat both disorders, is a growing area of research.

TBI sustained during combat is usually associated with an explosion, such as from artillery or an improvised explosive device, and these blasts often propel soldiers against a wall or the interior of a vehicle, which can result in a deceleration head injury. Although differentiation between the injury from the blast and the deceleration head trauma might not always be possible, investigations are under way not only to track severity of head trauma in soldiers

by using accelerometers in 'smart helmets', but also to potentially counteract harmful effects of the blast.

Long-term sequelae

Cause and effect relationships for the delayed sequelae of single-incident severe TBI are not well understood, owing to the variable latent period between injury and subsequent neurological and neuropsychiatric dysfunction. Some closed-head injuries involve only brief loss of consciousness—that is, mild TBI—but memory, affective and executive dysfunction can nevertheless emerge and cause substantial impairment and life disruption. Even when no overt damage is apparent on neuroimaging, such symptoms can linger for months, causing disruption of learning with subsequent poor academic or work performance.

Severe single-incident TBI, with or without skull fracture, can lead to permanent brain damage, with incomplete recovery and residual sensory, motor and cognitive deficits. Unlike mild repetitive TBI, discussed below, severe single-incident TBI is associated with increased risk of late-onset Alzheimer disease (AD).^{15–20} As the late consequences of TBI generate pathology that is reminiscent of AD or pure tauopathy, we posit that these two disorders have a shared pathogenesis. However, we must keep in mind that the pathogenesis of spontaneous or genetic AD or tauopathy *per se* might not be equivalent to that of post-traumatic neurodegeneration.

A recent study found that some neurodegenerative causes of death (namely, AD and amyotrophic lateral sclerosis) were four times higher in US National Football League players than in the general population.²¹ No such trend in the prevalence of Parkinson disease was noted. Paradoxically, the overall mortality rate of these retired players was substantially lower than that of the general population. Independent replication of the study should help to clarify this puzzling dissociation of outcomes. Notably, differential diagnosis of AD versus CTE on clinical grounds is not entirely reliable, which highlights a key knowledge gap in this area: no compelling epidemiological data are available to enable estimation of the incidence or prevalence of these neurodegenerative sequelae of TBI. Differential diagnosis in the future is likely to improve with the advent of neuropathology imaging—that is, amyloid imaging, which is already in clinical use,^{22,23} and tauopathy imaging, which is showing promise^{24,25} but is still in development.

Sequence of pathological events

Extrapolating from the known pathological and clinical course of AD and other tauopathies, we can construct a hypothetical temporal sequence of events in TBI and CTE. The most frequently proposed mechanism in TBI is 'diffuse axonal injury' (Figure 1), which is associated with alterations in many physiological processes, and accumulation of abnormal protein aggregates in cells and the brain parenchyma.² Altered proteolytic and proteostatic pathways are associated with changes that are evident at the histological level. Here, the similarity between pathways of idiopathic and post-traumatic neurodegeneration is most evident, in that both involve accumulation of apparently identical protein aggregates.

Within hours of single-incident severe TBI in humans and experimental animals, dramatic accumulation of amyloid precursor protein is evident in damaged axons and cell bodies.²⁶ Some studies in case series demonstrated that these changes are associated with increased brain concentrations of soluble amyloid- β_{42} ($A\beta_{42}$) peptides and deposition of diffuse $A\beta$ plaques in brain tissue in approximately 30% of patients with severe TBI, regardless of patient age (Figure 2).^{27–29} These plaques, which are predominantly $A\beta_{42}$ -immunoreactive, are similar to those observed in the earliest stages of AD pathology. An association between densities of such deposits and the apolipoprotein E (*APOE*) $\epsilon 4$ allele, which is in itself a risk factor for AD, has been established in boxers^{30,31} and patients with fatal head injuries.³² In addition, polymorphisms in the gene encoding neprilysin, an $A\beta$ -degrading enzyme, probably influence the rate of $A\beta$ degradation and clearance after TBI.³³

Some patients with parkinsonism have a history of single-incident TBI.^{34,35} Genetic, environmental and physical factors presumably distinguish people who are destined to develop parkinsonism from those destined to manifest AD years after TBI. Notably, precise dose–response data linking injury severity with induction of pathology in sports associated with neurodegeneration are not available. Similarly, no sport-specific factors are known that can distinguish TBI associated with boxing versus American football, soccer, hockey and wrestling. Typically, TBI associated with these sports is combined under the rubric of ‘mild repetitive’ TBI.

Mild repetitive TBI

The classic clinicopathological correlation linking neuro cognitive effects of repetitive mild head injury with tauopathy was described in boxers by Corsellis.^{36,37} The pathology is distinct from the clinical and pathological sequelae of severe single-incident TBI. Landmark neuro pathology studies reported that dementia pugilistica, or ‘punch drunk’ syndrome, involves prominent tauopathy, with patchy distribution of neurofibrillary tangles (NFTs) and neuropil threads throughout the neocortex.^{36–39} Diffuse $A\beta$ plaques were present in 50–100% of cases depending on the series, whereas tauopathy was a universal finding across all case series.^{38,40}

Dementia pugilistica

Dementia pugilistica typically involves NFT accumulation in the superficial grey matter, especially at the base of the sulci, with prominent perivascular and periventricular pathology. Subsequent immunohistochemistry studies of the dementia pugilistica cases that were originally examined by Corsellis demonstrated that all cases had diffuse $A\beta$ deposits (but not neuritic plaques) in addition to NFTs.^{38,39} The precise relationships between these lesions and clinical symptoms of dementia pugilistica is unknown, but may involve axonal injury, as traumatic tearing of neuronal connections (‘axonal shearing’, a component of diffuse axonal injury) is known to impair cortical and thalamic circuitry, thereby contributing to cognitive impairment and dementia.

As with single-incident TBI, a role for genetic factors in the development of AD neuropathology and dementia following mild repetitive TBI is emerging.^{30,41} The *APOE* $\epsilon 4$ allele is a well-established risk factor for amyloid plaque deposition and AD.^{42,43} Boxers

who were *APOE* $\epsilon 4$ carriers were more likely to develop dementia pugilistica than those lacking the $\epsilon 4$ allele,³⁰ suggesting that this allele confers greater vulnerability to brain pathology in trauma. Parkinsonism is also associated with dementia pugilistica. When rigidity and tremor dominate the clinical picture, the term 'pugilistic parkinsonism' is applied. An abundance of NFTs and the absence of Lewy bodies distinguishes pugilistic parkinsonism from idiopathic Parkinson disease. However, as in Parkinson disease, nigral neurons are lost in pugilistic parkinsonism, which probably underlies the clinical symptoms of this syndrome, although the precise mechanism is not known.

Chronic traumatic encephalopathy

In addition to studies in boxers, examination of the brains of several professional American football players and wrestlers has revealed an explanation for the little-discussed fact that football players who played for many years often suffered cognitive and neuropsychiatric decline in later life. Retired players with three or more reported concussions were found to have fivefold increased prevalence of mild cognitive disorders and threefold increased prevalence of substantial memory problems compared with healthy controls.⁴⁴ Although later cognitive decline in long-time professional football players had been noted anecdotally for years, the first autopsy report on such a player appeared in the literature only recently.⁴⁵ The pathology was indistinguishable from that of dementia pugilistica. This case had frequent diffuse neocortical A β plaques and sparse NFT pathology.⁴⁵ To distinguish the aetiology from that of boxing, Omalu and DeKosky reinstated the term 'chronic traumatic encephalopathy',^{46,47} a term from the 1920s and 1950s that caught on with the popular press in the contexts of sports and military injury.

CTE is characterized by distinct patterns of A β and tau pathology, although the extent of pathology varies between CTE cases (Figures 2 and 3). McKee and colleagues⁴⁸ noted that nearly 50% of CTE cases selected from the literature (on basis of the presence of NFT pathology) also had diffuse A β plaques, with nearly 30% of cases having classic AD (neuritic) plaques. Whereas boxing scoring includes records of knockouts, concussion histories in football players have been notoriously poor, owing to both absence of meticulous records and the culture of the sport, in which injuries are ignored by both players and coaching staff, to keep players in the game. The index case report of TBI in American football⁴⁵ has since been followed by confirmatory case studies,^{49–52} which found that cognitive decline began years after cessation of play.

Cohort composition and player position might influence assessment and determination of the risk, diagnosis and mechanisms of CTE. In the 'early days' of professional football, players wore leather helmets, which were later replaced by plastic helmets with canvas strap-interiors and foam cushioning inside. Modern US football helmets, including face masks, have not prevented concussions or severe TBI with loss of consciousness. Modern technology involving 'smart helmets' that record the physics of impact will help us to understand and quantify such injuries.

Dementia pugilistica had been recognized for decades, whereas reports of a similar phenomenon in football emerged in the late 1990s and early 2000s. At the Professional Football Hall of Fame, long-standing players would gather every year, providing a unique

opportunity to see high-level players from year to year. As the former players aged, increasing numbers of their members were noted to have memory problems. In addition to legal claims against the National Football League for the obvious orthopaedic injuries from playing football, claims for neurocognitive damage also began to emerge. One of the early players to sue for cognitive and neuropsychiatric injury was the first professional football player to undergo autopsy and can be regarded as the index case of sports-related CTE.^{44,45,53}

Once the problem was identified, researchers and the National Football League, as well as other athletic organizing bodies, responded rapidly. Since the emergence of multiple cases from several independent pathology laboratories and clinics,^{40,47} and in other high-impact sports such as wrestling,⁵¹ the phenomenon has been accepted by the medical and athletic authorities, with appropriate attempts to minimize its occurrence in contact sports. At least 12 former National Football League players have committed suicide over the past 25 years, many suffering from cognitive and affective symptoms. In two of the more recent suicides, the players shot themselves in the heart, which enabled their brains to be studied postmortem.^{53,55,56}

Broad public awareness of CTE now exists. We have little idea, however, of the risk of developing CTE following TBI, or of the basis of interindividual differences in susceptibility to this disorder. *APOE* $\epsilon 4$ alleles³⁰ might increase the risk of CTE in professional football players as they do in boxing, but conclusive evidence is lacking. Polymorphisms in the A β -degrading enzyme neprilysin have also been provisionally linked to CTE.³³ The largest collected reports of CTE have not published genetic analyses. Coordinated national and international approaches to CTE research are needed.

CTE research centres could be modelled on the existing NIH Alzheimer Disease Centers and the Alzheimer Disease Neuroimaging Initiative (ADNI). Indeed, the US Department of Defense has recently initiated a 'military TBI ADNI', and the National Football League has teamed up with the NIH to launch new research into CTE. An important starting point will be lifelong TBI diary databases for those at highest risk for TBI and CTE, in order to obtain reliable estimates of the lifetime accumulation of TBI and lifetime risk of developing CTE. Not every boxer develops dementia pugilistica and not every football player develops CTE; given the current state of technology, whole-genome sequencing of patients with CTE could provide comprehensive genetic profiles of those who develop CTE as well as those who experience identical lifelong TBI histories but remain cognitively intact.

Many soldiers experience head trauma periodically in training. Omalu recently reported the index case of 'military CTE' in a veteran of the Iraq war who committed suicide,⁵⁷ and suggested a link to the clinical PTSD phenotype, as mentioned above. These observations were recently confirmed and extended by an independent group of investigators.⁵⁸

Biomarkers

Neuroimaging

Increasing evidence suggests that single-incident severe TBI and mild repetitive concussions are associated with a heterogeneous profile of neuropathological changes resembling those in chronic neurodegenerative disorders, including AD and Parkinson disease. Such a similarity supports the use of amyloid-binding PET ligands for *in vivo* monitoring of neuropathology after TBI. PET ligands for imaging of tauopathy and synucleinopathy are currently in development (Figure 4). Clinical diagnosis of AD can be aided by quantifying A β , tau, and phospho-tau levels in CSF,⁵⁹ even in the absence of a positive amyloid PET scan.⁶⁰ These biomarkers could be especially useful for differential diagnosis of CTE,⁶¹ as the A β deposits are primarily diffuse, and NFT distribution is different to that in AD and frontotemporal dementia. Prior to regulatory approval, these ligands must be validated against neuropathology to ensure that the imaging signal corresponds to the underlying amyloid or tau pathology, as already demonstrated for Pittsburgh compound B⁶² and florbetapir.²³

Body fluid biomarkers

The clinical diagnosis and evaluation of single-incident severe TBI, which involves the combination of clinical examination and MRI, is relatively straightforward. A substantial challenge for the clinician, however, is assessment of whether patients with milder forms of head trauma, and nonspecific symptoms such as dizziness and headache, have acquired permanent damage to the brain. Indeed, mild TBI can cause neuronal damage with selective swelling and disconnection of long axons in the white matter of the brain.⁶³ Such challenges can be addressed by measurement of biochemical markers in body fluids, such as serum, plasma, urine or CSF.

Cerebrospinal fluid biomarkers

For brain disorders such as TBI, the advantage of CSF biomarkers is that the CSF is in direct contact with the brain extracellular fluid, which mirrors biochemical changes in the brain. As reviewed in another article in this issue, the most promising CSF biomarkers for acute TBI are axonal proteins, such as neurofilament light polypeptide and tau.⁶¹ The CSF levels of these proteins probably reflect the extent of axonal damage after acute TBI. Indeed, a marked increase in CSF total tau that correlates with clinical outcome is found in patients after severe TBI.^{64,65}

These biomarkers could be especially important in the evaluation of individuals with mild TBI such as boxers, who endure repeated TBI by head punches during sparring or bouts. Interestingly, a longitudinal study of amateur boxers showed a marked increase in levels of CSF neurofilament light polypeptide, together with a less pronounced increase in tau, after bouts.⁶⁶ The increase in neurofilament light polypeptide was much more pronounced in boxers who sustained several head punches. These biomarker changes seem to be transitory, as CSF neurofilament light polypeptide levels returned to normal levels after a 3-month period of rest from bouts and sparring.⁶⁷ Similar results have been reported in an independent study on Olympic boxers, confirming a direct correlation between increases in

CSF biomarker levels and boxing exposure.⁶⁸ These findings suggest that CSF levels of neurofilament light polypeptide and total tau could be valuable biomarkers for monitoring of axonal damage in boxers (Figure 5) and other TBI cases.

Plasma biomarkers

Given that CSF samples must be obtained by invasive lumbar puncture, availability of biomarkers of brain damage that can be assayed in blood samples would be beneficial. Several protein biomarkers have been examined as potential blood markers of TBI.⁶¹ Of these, serum levels of the glial protein S-100B increased markedly in patients with severe TBI and correlated with clinical outcome.^{69,70} Reports are inconsistent, however, regarding the correlation between serum S-100B levels and intra cranial pathology as evaluated by CT or MRI scans, or clinical outcome in patients with mild TBI.^{69,70} A pilot study found an increase in plasma tau to more than 300% of control levels in amateur boxers following a bout.⁷¹ Tau levels in blood samples might, therefore, serve as a biomarker of mild TBI.

Several factors could contribute to the lack of plasma biomarkers of TBI: first, dilution of the brain-specific protein, both in the large volume of plasma and in the extracellular fluid of peripheral organs; second, degradation of the biomarker candidate by blood proteases; and third, clearance of the protein by hepatic metabolism or renal excretion. Furthermore, analyses of brain proteins in blood can be confounded by release of the same protein from peripheral tissues. For example, S-100B is released from fractured bones and injured skeletal muscles in patients with multiple trauma, thereby reducing the diagnostic power of this marker for TBI.^{69,70} Highly sensitive analytical techniques are needed to detect minute amounts of brain-specific proteins in peripheral blood as biomarkers for mild TBI. Measurement of tau protein in serum samples using a novel ultra-sensitive technique called digital ELISA, which involves single-molecule arrays, has recently been described.⁷² This assay has a limit of detection of 0.02 pg/ml, which is 1000-fold more sensitive than conventional immunoassays, and showed good performance in identifying brain damage in patients who had brain hypoxia following cardiac arrest.⁷² Studies are ongoing to examine whether serum levels of tau could serve as a blood biomarker of mild TBI.

Biomarker applications

Clinical studies of TBI biomarkers in professional contact sports such as boxing and martial arts, American foot ball, ice hockey and rugby, as well as in military personnel at risk of blast overpressure and direct traumatic head injuries, would be particularly valuable. Clinical evaluation and biomarker examination should ideally be performed at baseline (for example, before the football season or before military service in a combat zone) and after TBI. Such studies would provide important information both on pathogenic mechanisms in acute TBI and on the threshold of exposure that is needed to induce detectable neuronal damage.

CSF and other biomarkers of acute TBI could be important tools for medical counselling of concussed athletes and other risk groups, such as military personnel, providing both diagnostic information and prognostic guidance. Post-injury CSF biomarker levels might give information on the severity of axonal damage after a knockout or blast injury, and

follow-up biomarker testing could be used to monitor when active brain-cell lysis has resolved—that is, when CSF biomarker levels return to normal—and, thereby, when athletes can return to their sport, or military personnel to service. Much research is needed before we come to this point in the medical care of patients with acute TBI. The ADNI study, mentioned above, provides an outstanding example of a multicentre, longitudinal study to understand pathogenic mechanisms, their interrelationships and their temporal evolution in AD, which could provide a useful model for future TBI research.⁷³ These studies would not only provide data on the incidence and prevalence of chronic brain disorders, but also improve our understanding of the relationship between acute and long-term sequelae of repetitive TBI.

During life, clinical differentiation between delayed post-traumatic AD and CTE remains a challenge. CSF biomarker analysis or amyloid PET data are not currently available for autopsy-confirmed CTE cases, although the first case report of a negative amyloid scan in a retired National Football League player with the clinical diagnosis of probable CTE has recently been published.⁷⁴ Current amyloid PET ligands detect fibrillar A β plaques, whereas imaging the diffuse A β deposits that are often part of CTE can be challenging. Advances have recently been made in PET imaging of tau, and accumulation of clinical experience with this imaging tool is expected to begin in 2013.^{24,25,75}

Biomarkers would provide information on pathogenic mechanisms and the temporal evolution of different forms of pathology, which would also help to resolve the question of whether pathogenic mechanisms differ between dementia pugilistica and CTE. The temporal-sequence information will be highly informative for development and timing of medical interventions. Lastly, these studies would be important for formulation of widely accepted, evidence-based guidelines for the clinical diagnosis of CTE.

Regulation of contact sports

No treatment is available for dementia pugilistica or CTE, but these conditions—in contrast to most other neurodegenerative disorders—are preventable. In 2005, the World Medical Association (WMA) recommended banning boxing⁷⁶ because of the basic intent of the sport to inflict bodily harm to the opponent. Several alternative ways to make the sport safer exist, including measures to reduce the number of head punches during a bout, use of protective equipment to limit the deleterious effects on the brain of repeated punches, and various approaches to prevent boxers from continuing a potentially dangerous fight, including education and action of the referees.

The tragic death of the professional boxer Kim Duk-Koo in 1982 owing to a subdural haematoma after being knocked out by Ray Mancini in round 14 served as an initiating factor to lower the number of rounds in professional boxing from 15 to 12. Following this change, a trend toward decreased mortality amongst professional boxers has been observed, although this trend might also be related to lower exposure to repetitive head trauma in this group of individuals, with shorter careers and fewer fights.⁷⁷

Impact forces from punches have been studied experimentally, and can be comparable to the impact of a 6-kg bowling ball rolling at 20 mph.⁷⁸ Boxing helmets and gloves reduce the

impact from a punch.⁷⁹ Protective headgear is mandatory in amateur boxing but not in professional boxing or mixed martial arts—a sport that is rising in popularity. Although helmets in sports reduce incidence of superficial facial injury and severe head injury, no clear evidence is available to show that they reduce the risk of concussion.^{80,81} Moreover, although protective equipment might reduce risk of CTE in amateur boxing, knockouts still occur, and around 20% of deaths due to brain damage occur in amateur boxing.⁸² Nevertheless, introduction of thicker gloves and headgear with thicker padding—as recommended by the WMA⁸³—would probably reduce the risk of CTE and concussions.

Several international sports organizations, including the International Olympic Commission (IOC), the Fédération Internationale de Football Association, the International Rugby Board, and the International Ice Hockey Federation, have worked on consensus guidelines on sports-related concussion.⁸¹ These guidelines state that an athlete who shows any symptoms of concussion should be removed from play immediately and evaluated by a physician, and should not be allowed to return to play until symptoms have subsided.^{82–84} As the aim of boxing and martial arts sports is to inflict the opponent acute brain damage that is severe enough to win the bout by knockout, such guidelines are difficult to implement in these sports. Given that amateur boxing is an Olympic sport, this issue could present a delicate problem for the IOC.

Conclusions and future directions

The utility of the diagnosis of CTE remains to be studied and improved. Some researchers have questioned the identity of dementia pugilistica and CTE, given the apathetic clinical picture of the former in contrast to the frequently violent clinical picture of the latter (P. Davies, personal communication). Some individuals involved in organized sport remain unconvinced of the potential seriousness of the problem.⁸⁵

A β plaques are more common in CTE than in acute TBI. The main distinguishing features of CTE are the disproportionate tauopathy relative to the amyloidosis and the concentration of CTE lesions at the base of the gyri. Research on TBI and CTE is aimed at elucidating the neuropathological basis of these differences.⁶³ Laboratory modelling of TBI and CTE should facilitate elucidation of the cellular and molecular changes underlying these phenomena. The brain–skull–spine configurations of current rodent and swine models, however, are limited in their capacity to reflect the situation in humans, calling for design of improved models in other species. Despite this challenge, in the meantime, genetically modified rodent models provide powerful tools for delineation of TBI pathways. Several rodent models of CTE have been reported,^{58,86–93} but have yet to be confirmed. Using AD as a model for CTE progression, the acute phase is predicted to be largely A β -related, whereas the chronic phase may be more A β -independent and tauopathy-driven. Preliminary molecular pathology and experimental pharmacology data, however, suggest that at least some features of the AD model might or might not apply to TBI and CTE.^{87–91} Clearly, more research is required.

The incidence and prevalence of CTE and the role of genetic factors remains to be elucidated.⁹⁴ Long-term follow-up of multiple cohorts of individuals, such as American

football players and soldiers, who have sustained repetitive injuries of undefined number and severity, will be valuable for determination and replication of environmental, genetic and other risk factors that influence the natural history of TBI and CTE. TBI diaries will be helpful in determining the number and severity of head injuries, to allow estimation of cumulative risk of TBI to athletes.

Major clinical⁹⁵ and neuroimaging⁹⁶ efforts are under way, and these programmes will be facilitated by recent plans to place neurologists on the sidelines at National Football League games.⁹⁷ These data can serve as models for prospective studies of the cognitive, neuropsychiatric and motor performance of soldiers, athletes and other exposed populations, and can inform behavioural and pharmacological interventions aimed at prophylaxis and therapy.

A future challenge will be translation of our improved understanding of TBI and CTE pathogenesis into rational, evidence-based changes in regulation of sports by governments and school boards that will minimize the public's exposure to TBI and its chronic neurodegenerative sequelae. In addition, the pathological cascades in TBI and CTE, in both acute and chronic stages, must be understood to enable identification of valid biomarkers and development of treatments.

Acknowledgments

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References

- Centers for Disease Control and Prevention. Injury prevention and control: traumatic brain injury. Centers for Disease Control and Prevention; 2012. [online], <http://www.cdc.gov/traumaticbraininjury/statistics.html>
- DeKosky ST, Ikonomic MD, Gandy S. Traumatic brain injury—football, warfare, and long-term effects. *N. Engl. J. Med.* 2010; 363:1293–1296. [PubMed: 20879875]
- US Department of Defense. Traumatic brain injury—Department of Defense special report. US Department of Defense; 2012. [online], http://www.defense.gov/home/features/2012/0312_tbi/
- Lange, RT.; Brickell, TA.; Ivins, B.; Vanderploeg, R.; French, LM. Variable, not always persistent, postconcussion symptoms following mild TBI in U.S. military service members: a 5-year cross-sectional outcome study.. *J. Neurotrauma.* <http://dx.doi.org/10.1089/neu.2012.2743>
- Wall PL. Posttraumatic stress disorder and traumatic brain injury in current military populations: a critical analysis. *J. Am. Psychiatr. Nurses Assoc.* 2012; 18:278–298. [PubMed: 23053745]
- Capehart B, Bass D. Review: managing posttraumatic stress disorder in combat veterans with comorbid traumatic brain injury. *J. Rehabil. Res. Dev.* 2012; 49:789–812. [PubMed: 23015586]
- Vasterling JJ, et al. Neuropsychological outcomes of mild traumatic brain injury, post-traumatic stress disorder and depression in Iraq- deployed US Army soldiers. *Br. J. Psychiatry.* 2012; 201:186–192. [PubMed: 22743844]
- Vanderploeg RD, et al. Health outcomes associated with military deployment: mild traumatic brain injury, blast, trauma, and combat associations in the Florida National Guard. *Arch. Phys. Med. Rehabil.* 2012; 93:1887–1895. [PubMed: 22705240]

10. Bazarian JJ, et al. The relation between posttraumatic stress disorder and mild traumatic brain injury acquired during operations enduring freedom and Iraqi freedom. *J. Head Trauma Rehabil.* 2013; 28:1–12. [PubMed: 22647965]
11. Shively SB, Perl DP. Traumatic brain injury, shell shock, and posttraumatic stress disorder in the military—past, present, and future. *J. Head Trauma Rehabil.* 2012; 27:234–239. [PubMed: 22573042]
12. Ruff RL, Riechers RG 2nd, Wang XF, Piero T, Ruff SS. A case–control study examining whether neurological deficits and PTSD in combat veterans are related to episodes of mild TBI. *BMJ Open.* 2012; 2:e000312.
13. Barnes SM, Walter KH, Chard KM. Does a history of mild traumatic brain injury increase suicide risk in veterans with PTSD? *Rehabil. Psychol.* 2012; 57:18–26. [PubMed: 22369114]
14. Bogdanova Y, Verfaellie M. Cognitive sequelae of blast-induced traumatic brain injury: recovery and rehabilitation. *Neuropsychol. Rev.* 2012; 22:4–20. [PubMed: 22350691]
15. Moretti L, et al. Cognitive decline in older adults with a history of traumatic brain injury. *Lancet Neurol.* 2012; 11:1103–1112. [PubMed: 23153408]
16. Shively S, Scher AI, Perl DP, Diaz-Arrastia R. Dementia resulting from traumatic brain injury: what is the pathology? *Arch. Neurol.* 2012; 9:1–7.
17. Sivanandam TM, Thakur MK. Traumatic brain injury: a risk factor for Alzheimer's disease. *Neurosci. Biobehav. Rev.* 2012; 36:1376–1381. [PubMed: 22390915]
18. Esiri MM, Chance SA. Cognitive reserve, cortical plasticity and resistance to Alzheimer's disease. *Alzheimers Res. Ther.* 2012; 4:7. [PubMed: 22380508]
19. Norton MC, et al. Lifestyle behavior pattern is associated with different levels of risk for incident dementia and Alzheimer's disease: the Cache County study. *J. Am. Geriatr. Soc.* 2012; 60:405–412. [PubMed: 22316091]
20. Johnson VE, Stewart W, Smith DH. Widespread τ and amyloid- β pathology many years after a single traumatic brain injury in humans. *Brain Pathol.* 2012; 22:142–149. [PubMed: 21714827]
21. Lehman EJ, Hein MJ, Baron SL, Gersic CM. Neurodegenerative causes of death among retired National Football League players. *Neurology.* 2012; 79:1970–1974. [PubMed: 22955124]
22. Klunk WE, et al. Imaging brain amyloid in Alzheimer's disease with Pittsburgh Compound-B. *Ann. Neurol.* 2004; 55:306–319. [PubMed: 14991808]
23. Clark CM, et al. Use of florbetapir-PET for imaging β -amyloid pathology. *JAMA.* 2011; 305:275–283. [PubMed: 21245183]
24. Zhang W, et al. A highly selective and specific PET tracer for imaging of tau pathologies. *J. Alzheimers Dis.* 2012; 31:601–612. [PubMed: 22683529]
25. Chien, DT., et al. Early clinical PET imaging results with the novel PHF-tau radioligand [F-18]-T807. *J. Alzheimers Dis.* <http://dx.doi.org/10.3233/JAD-122059>
26. Ciallella JR, et al. Changes in expression of amyloid precursor protein and interleukin-1 β after experimental traumatic brain injury in rats. *J. Neurotrauma.* 2002; 19:1555–1567. [PubMed: 12542857]
27. Ikonovic MD, et al. Alzheimer's pathology in human temporal cortex surgically excised after severe brain injury. *Exp. Neurol.* 2004; 190:192–203. [PubMed: 15473992]
28. DeKosky ST, et al. Association of increased cortical soluble A β ₄₂ levels with diffuse plaques after severe brain injury in humans. *Arch. Neurol.* 2007; 64:541–544. [PubMed: 17420316]
29. Nicoll JA, Roberts GW, Graham DI. Apolipoprotein E ϵ 4 allele is associated with deposition of amyloid β -protein following head injury. *Nat. Med.* 1995; 1:135–137. [PubMed: 7585009]
30. Jordan BD, et al. Apolipoprotein E ϵ 4 associated with chronic traumatic brain injury in boxing. *JAMA.* 1997; 278:136–140. [PubMed: 9214529]
31. Jordan, BD. Clinical spectrum of sports-related traumatic brain injury. *Nat. Rev. Neurol.* <http://dx.doi.org/10.1038/nrneurol.2013.33>
32. Horsburgh K, et al. β -amyloid (A β)₄₂(43), a β ₄₂, a β ₄₀ and apoE immunostaining of plaques in fatal head injury. *Neuropathol. Appl. Neurobiol.* 2000; 26:124–132. [PubMed: 10840275]
33. Johnson VE, et al. A neprilysin polymorphism and amyloid- β plaques after traumatic brain injury. *J. Neurotrauma.* 2009; 26:1197–1202. [PubMed: 19326964]

34. Factor SA, Weiner WJ. Prior history of head trauma in Parkinson's disease. *Mov. Disord.* 1991; 6:225–229. [PubMed: 1922127]
35. Rugbjerg K, Ritz B, Korbo L, Martinussen N, Olsen JH. Risk of Parkinson's disease after hospital contact for head injury: population based case–control study. *BMJ.* 2008; 337:a2494. [PubMed: 19074944]
36. Corsellis JA, Brierley JB. Observations on the pathology of insidious dementia following head injury. *J. Ment. Sci.* 1959; 105:714–720. [PubMed: 13812088]
37. Corsellis JA. Boxing and the brain. *BMJ.* 1989; 298:105–109. [PubMed: 2493277]
38. Roberts GW, Allsop D, Bruton C. The occult aftermath of boxing. *J. Neurol. Neurosurg. Psychiatry.* 1990; 53:373–378. [PubMed: 2191084]
39. Tokuda T, Ikeda S, Yanagisawa N, Ihara Y, Glenner GG. Re-examination of ex-boxers' brains using immunohistochemistry with antibodies to amyloid β -protein and tau protein. *Acta Neuropathol.* 1991; 82:280–285. [PubMed: 1759560]
40. McKee AC, et al. Chronic traumatic encephalopathy in athletes: progressive tauopathy after repetitive head injury. *J. Neuropathol. Exp. Neurol.* 2009; 68:709–735. [PubMed: 19535999]
41. Gandy S, DeKosky ST. APOE ϵ 4 status and traumatic brain injury on the gridiron or the battlefield. *Sci. Transl. Med.* 2012; 4:134ed4.
42. Gandy S. The role of cerebral amyloid β accumulation in common forms of Alzheimer disease. *J. Clin. Invest.* 2005; 115:1121–1129. [PubMed: 15864339]
43. Gandy S, DeKosky ST. Toward the treatment and prevention of Alzheimer's disease: rational strategies and recent progress. *Annu. Rev. Med.* 2013; 64:367–383. [PubMed: 23327526]
44. Guskiewicz KM, et al. Association between recurrent concussion and late-life cognitive impairment in retired professional football players. *Neurosurgery.* 2005; 57:719–726. [PubMed: 16239884]
45. Omalu BI, et al. Chronic traumatic encephalopathy in a National Football League player. *Neurosurgery.* 2005; 57:128–134. [PubMed: 15987548]
46. Yi J, Padalino DJ, Chin LS, Montenegro P, Cantu RC. Chronic traumatic encephalopathy. *Curr. Sports Med. Rep.* 2013; 12:28–32. [PubMed: 23314081]
47. Clark EC, Harper EO. Electroencephalographic findings in 186 cases of chronic post traumatic encephalopathy. *Electroencephalogr. Clin. Neurophysiol.* 1951; 3:9–14.
48. McKee AC, et al. The spectrum of disease in chronic traumatic encephalopathy. *Brain.* 2013; 136:43–64. [PubMed: 23208308]
49. Omalu BI, Hamilton RL, Kamboh MI, DeKosky ST, Bailes J. Chronic traumatic encephalopathy (CTE) in a National Football League Player: case report and emerging medicolegal practice questions. *J. Forensic Nurs.* 2010; 6:40–46. [PubMed: 20201914]
50. Omalu BI, Bailes J, Hammers JL, Fitzsimmons RP. Chronic traumatic encephalopathy, suicides and parasuicides in professional American athletes: the role of the forensic pathologist. *Am. J. Forensic Med. Pathol.* 2010; 31:130–132. [PubMed: 20032774]
51. Omalu BI, Fitzsimmons RP, Hammers J, Bailes J. Chronic traumatic encephalopathy in a professional American wrestler. *J. Forensic. Nurs.* 2010; 6:130–136. [PubMed: 21175533]
52. Omalu B, et al. Emerging histomorphologic phenotypes of chronic traumatic encephalopathy in American athletes. *Neurosurgery.* 2011; 69:173–183. [PubMed: 21358359]
53. Dwyre, B. Dave Duerson's suicide could be a turning point for the NFL. *Los Angeles Times*; Feb 21. 2011 <http://articles.latimes.com/2011/feb/21/sports/la-sp-dwyre-20110222>
54. Nelson, E.; Sherwood, R. Chris Benoit's murder, suicide: was brain damage to blame?. *ABC News*; Aug 26. 2010 <http://abcnews.go.com/Nightline/chris-benoits-dad-son-suffered-severe-brain-damage/story?id=11471875>
55. Tierny, M. Football player who killed himself had brain disease. *Vol. B16. New York Times*; Jul 27. 2012
56. Kounang, N.; Smith, S. Seau had brain disease that comes from hits to head, NIH finds. *CNN*; Jan 11. 2013 <http://www.cnn.com/2013/01/10/health/seau-brain-disease/index.html>
57. Omalu B, et al. Chronic traumatic encephalopathy in an Iraqi war veteran with posttraumatic stress disorder who committed suicide. *Neurosurg. Focus.* 2011; 31:E3. [PubMed: 22044102]

58. Goldstein LE, et al. Chronic traumatic encephalopathy in blast-exposed military veterans and a blast neurotrauma mouse model. *Sci. Transl. Med.* 2012; 14:134ra60.
59. Fagan AM, et al. Cerebrospinal fluid tau/ β -amyloid₄₂ ratio as a prediction of cognitive decline in nondemented older adults. *Arch. Neurol.* 2007; 64:343–349. [PubMed: 17210801]
60. Cairns NJ, et al. Absence of Pittsburgh compound B detection of cerebral amyloid β in a patient with clinical, cognitive, and cerebrospinal fluid markers of Alzheimer disease: a case report. *Arch. Neurol.* 2009; 66:1557–1562. [PubMed: 20008664]
61. Zetterberg, H.; Smith, DH.; Blennow, K. Biomarkers of mild traumatic brain injury in cerebrospinal fluid and blood. *Nat. Rev. Neurol.* <http://dx.doi.org/10.1038/nrneurol.2013.9>
62. Ikonomovic MD, et al. Post-mortem correlates of in vivo PiB-PET amyloid imaging in a typical case of Alzheimer's disease. *Brain.* 2008; 131:1630–1645. [PubMed: 18339640]
63. Povlishock JT, Becker DP, Cheng CL, Vaughan GW. Axonal change in minor head injury. *J. Neuropathol. Exp. Neurol.* 1983; 42:225–242. [PubMed: 6188807]
64. Franz G, et al. Amyloid beta 1–42 and tau in cerebrospinal fluid after severe traumatic brain injury. *Neurology.* 2003; 60:1457–1461. [PubMed: 12743231]
65. Öst M, et al. Initial CSF total tau correlates with 1-year outcome in patients with traumatic brain injury. *Neurology.* 2006; 67:1600–1604. [PubMed: 17101890]
66. Zetterberg H, et al. Neurochemical aftermath of amateur boxing. *Arch. Neurol.* 2006; 63:1277–1280. [PubMed: 16966505]
67. Neselius S, et al. CSF-biomarkers in Olympic boxing: diagnosis and effects of repetitive head trauma. *PLoS ONE.* 2012; 7:e33606. [PubMed: 22496755]
68. Kövesdi E, et al. Update on protein biomarkers in traumatic brain injury with emphasis on clinical use in adults and pediatrics. *Acta Neurochir. (Wien).* 2010; 152:1–17. [PubMed: 19652904]
69. Naeimi ZS, Weinhofer A, Sarahrudi K, Heinz T, Vecsei V. Predictive value of S-100B protein and neuron specific-enolase as markers of traumatic brain damage in clinical use. *Brain Inj.* 2006; 20:463–468. [PubMed: 16716992]
70. Anderson RE, Hansson LO, Nilsson O, Djalil-Merzoug R, Settergren G. High serum S100B levels for trauma patients without head injuries. *Neurosurgery.* 2001; 48:1255–1258. [PubMed: 11383727]
71. Neselius S, et al. Olympic boxing is associated with elevated levels of the neuronal protein tau in plasma. *Brain Inj.* (in press).
72. Randall, J., et al. Tau proteins in serum predict neurological outcome after hypoxic brain injury from cardiac arrest: results of a pilot study. *Resuscitation.* <http://dx.doi.org/10.1016/j.resuscitation.2012.07.027>
73. Weiner MW, et al. The Alzheimer's Disease Neuroimaging Initiative: a review of papers published since its inception. *Alzheimers Dement.* 2012; 8(Suppl. 1):S1–S68. [PubMed: 22047634]
74. Mitsis, E., et al. Florbetapir scanning excludes Alzheimer's disease in a retired NFL player with delayed cognitive impairment. Presented at the 7th Human Amyloid Imaging Conference; http://www.scribd.com/doc/117537451/HAI-2013-ConferenceBook#outer_page_83
75. Fodero-Tavoletti MT, et al. ¹⁸F-THK523: a novel in vivo tau imaging ligand for Alzheimer's disease. *Brain.* 2011; 134:1089–1100. [PubMed: 21436112]
76. World Medical Association. WMA statement on boxing. WMA; 2013. [online], <http://www.wma.net/en/30publications/10policies/b6/index.html>
77. Baird LC, et al. Mortality resulting from head injury in professional boxing. *Neurosurgery.* 2010; 67:1444–1450. [PubMed: 20948404]
78. Atha J, Yeadon MR, Sandover J, Parsons KC. The damaging punch. *Br. Med. J.* 1985; 291:1756–1757. [PubMed: 3936571]
79. Bartsch AJ, Benzel EC, Miele VJ, Morr DR, Prakash V. Boxing and mixed martial arts: preliminary traumatic neuromechanical injury risk analyses from laboratory impact dosage data. *J. Neurosurg.* 2012; 116:1070–1080. [PubMed: 22313361]
80. Jako P. Safety measures in amateur boxing. *Br. J. Sports Med.* 2002; 36:394–395. [PubMed: 12453832]

81. McCrory P, et al. Consensus statement on concussion in sport—the Third International Conference on Concussion in Sport held in Zurich, November 2008. *Phys. Sportsmed.* 2009; 37:141–159. [PubMed: 20048521]
82. Svinth, JR. Death Under the Spotlight: The Manuel Velazquez Boxing Fatality Collection.. *Electronic Journals of Martial Arts and Sciences.* 2007. [online], http://ejmas.com/jcs/jcsartsvinth_a_0700.htm
83. Miele VJ, Bailes JE. Objectifying when to halt a boxing match: a video analysis of fatalities. *Neurosurgery.* 2007; 60:307–315. [PubMed: 17290181]
84. Cantu RC. Return to play guidelines after a head injury. *Clin. Sports Med.* 1998; 17:45–60. [PubMed: 9475970]
85. Epstein, D. Conclusions? Too early.. *Sports Illustrated.* Jan 21. 2013 <http://sportsillustrated.cnn.com/vault/article/magazine/MAG1206738/index.htm>
86. Abrahamson EE, Ikonomic MD, Dixon CE, DeKosky ST. Simvastatin therapy prevents brain trauma-induced increases in β -amyloid peptide levels. *Ann. Neurol.* 2009; 66:407–414. [PubMed: 19798641]
87. Loane DJ, et al. Amyloid precursor protein secretases as therapeutic targets for traumatic brain injury. *Nat. Med.* 2009; 15:377–379. [PubMed: 19287391]
88. Tran HT, Sanchez L, Brody DL. Inhibition of JNK by a peptide inhibitor reduces traumatic brain injury-induced tauopathy in transgenic mice. *J. Neuropathol. Exp. Neurol.* 2012; 71:116–129. [PubMed: 22249463]
89. Magnoni S, et al. Tau elevations in the brain extracellular space correlate with reduced amyloid- β levels and predict adverse clinical outcomes after severe traumatic brain injury. *Brain.* 2012; 135:1268–1280. [PubMed: 22116192]
90. Tran HT, Sanchez L, Esparza TJ, Brody DL. Distinct temporal and anatomical distributions of amyloid- β and tau abnormalities following controlled cortical impact in transgenic mice. *PLoS ONE.* 2011; 6:e25475. [PubMed: 21980472]
91. Tran HT, LaFerla FM, Holtzman DM, Brody DL. Controlled cortical impact traumatic brain injury in 3xTg-AD mice causes acute intra- axonal amyloid- β accumulation and independently accelerates the development of tau abnormalities. *J. Neurosci.* 2011; 29:9513–9525. [PubMed: 21715616]
92. Garman RH, et al. Blast exposure in rats with body shielding is characterized primarily by diffuse axonal injury. *J. Neurotrauma.* 2011; 28:947–959. [PubMed: 21449683]
93. Ojo JO, et al. Repetitive mild traumatic brain injury augments tau pathology and glial activation in aged hTau mice. *J. Neuropathol. Exp. Neurol.* 2013; 72:137–151. [PubMed: 23334597]
94. Blennow K, Hardy J, Zetterberg H. The neuropathology and neurobiology of traumatic brain injury. *Neuron.* 2012; 76:886–899. [PubMed: 23217738]
95. Smith, S. NFL players association, Harvard planning \$100 million player study. *CNN*; Jan 30. 2013 <http://www.cnn.com/2013/01/29/health/nfl-harvard-study/index.html>
96. Battista, J. NFL joins with GE in effort to detect concussions. Vol. SP6. *New York Times*; Feb 3. 2013
97. NFL expects to soon have neurological consultants on sidelines.. *National Football League.* Jan 31. 2013 [No authors listed] [online], <http://www.nfl.com/news/story/0ap1000000133506/article/nfl-expects-to-soon-have-neurological-consultants-on-sidelines>

Key points

- Traumatic brain injury (TBI) can lead to delayed-onset neurodegenerative syndromes that include Alzheimer disease (AD) and chronic traumatic encephalopathy (CTE)
- CTE has gained attention owing to increasing media coverage of neuropsychiatric dysfunction in players of high-impact sport, such as boxing and American football
- Brain pathology after single-incident severe TBI is similar to early amyloid pathology in AD, whereas repetitive TBI can produce tauopathy with or without amyloidosis that resembles pathology of boxers' dementia
- Estimation of the risk and prevalence of CTE remains challenging, and accurate prediction of TBI outcome and CTE risk for soldiers and players of high-impact sports is not yet possible
- Several genetic risk factors for CTE have been proposed but remain to be established
- Cerebrospinal fluid and neuroimaging biomarkers of TBI and CTE are emerging and hold promise for antemortem diagnosis of CTE, prediction of CTE risk, and monitoring of neuropathology progression

Review criteria

Literature on dementia pugilistica and chronic traumatic brain injury published since 1950 was accessed via Internet-wide and database searches of both public and academic literature (that is, PubMed, PubMedCentral and MEDLINE). The search terms “brain trauma”, “brain injury”, and “dementia pugilistica” were used for broad searches, which were then narrowed by manual inspection. Reference lists from recent reviews of the topic were scanned for additional leads.

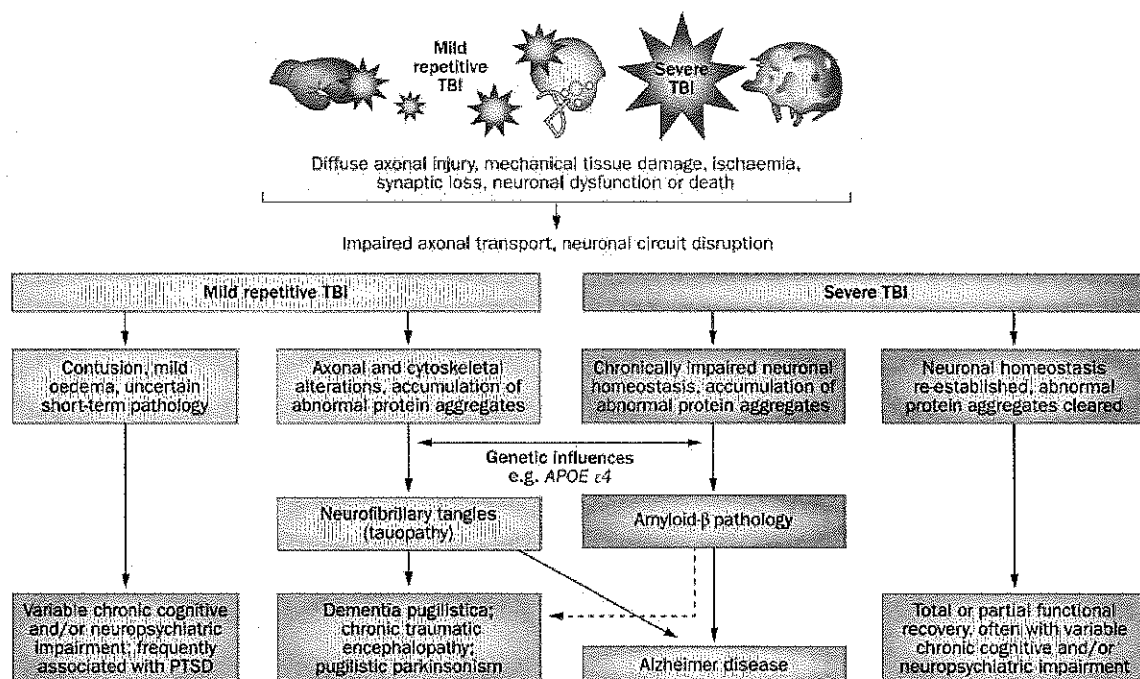


Figure 1.

Spectrum of pathological features and outcomes of mild and severe TBI. Abbreviations: *APOE*, apolipoprotein E; PTSD, post-traumatic stress disorder; TBI, traumatic brain injury.

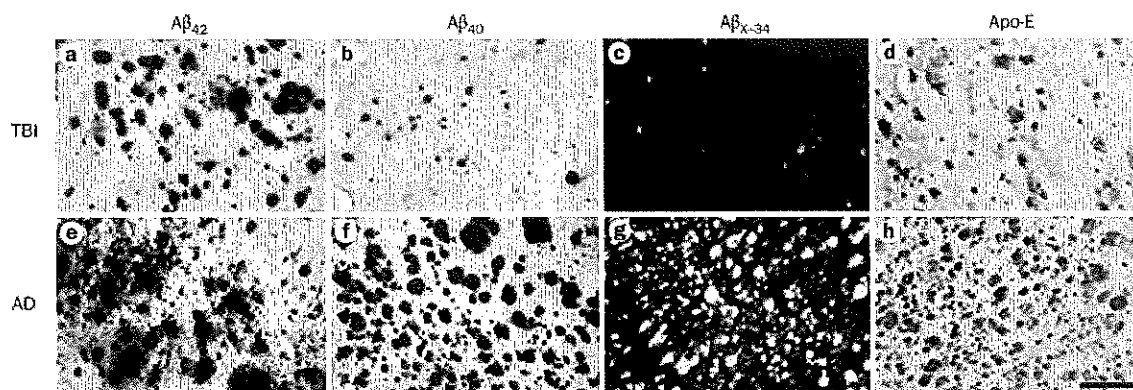


Figure 2.

Histopathology of the temporal cortex in TBI and AD. Brain sections from **a–d** | an individual with severe acute TBI and **e–h** | a patient with AD are labelled for A β ₄₂ (**a,e**), A β ₄₀ (**b,f**), A β _{X-34} (**c,g**), or Apo-E (**d,h**). In the TBI case, A β ₄₂ plaques (**a**) are more abundant than A β ₄₀ plaques (**b**). Frequency of plaques with A β _{X-34} (**c**) is closer to that of A β ₄₀, whereas distribution of Apo-E-positive plaques (**d**) is similar to A β ₄₂ plaques. The AD temporal cortex shows profusion of all plaque types (**e–h**), as well as neurofibrillary tangles, neuropil threads, and vascular amyloidosis (**g**). Scale bar: 200 μ m. Abbreviations: A β , amyloid- β ; AD, Alzheimer disease; Apo-E, apolipoprotein E; TBI, traumatic brain injury. Permission obtained from Elsevier Ltd © Ikonovic, M. D. *et al. Exp. Neurol.* **190**, 192–203 (2004).

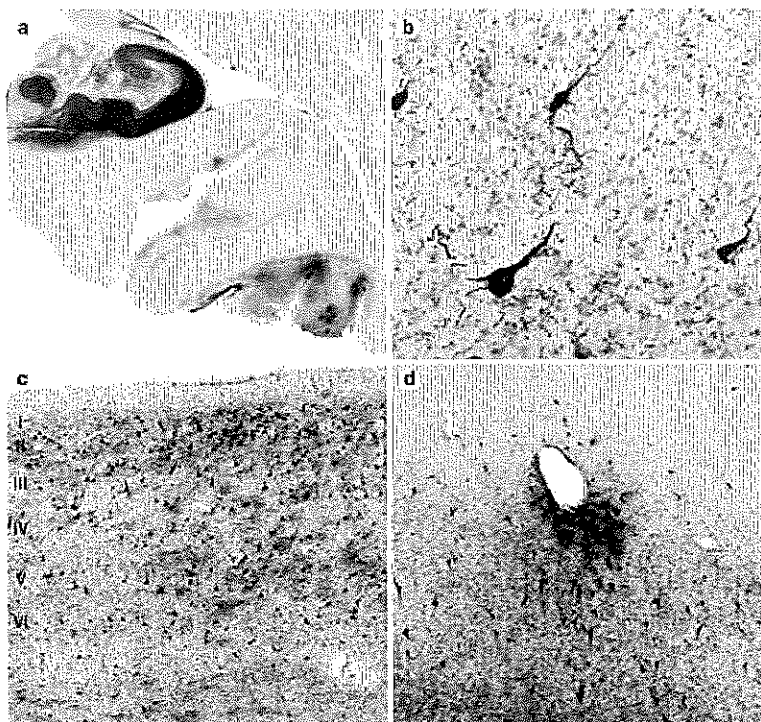


Figure 3.

Histopathological features of chronic traumatic encephalopathy in a former professional football player. All sections are immunostained for abnormally phosphorylated tau using an AT-8 monoclonal antibody that detects hyperphosphorylated tau (Ser202 and Thr205). **a** | Scanning view of the hippocampus and parahippocampal cortex. Note intense immunostaining of the entire Ammon horn and subiculum, with focal involvement at the depths of sulci of the inferior temporal lobe. Original magnification $\times 1$. **b** | Appearance of individual neurofibrillary tangles in the neocortex. Original magnification $\times 160$. **c** | Neurofibrillary tangles in the anterior insular cortex form preferentially in the superficial layers (layers II–III), rather than in deeper layers (layers V–VI) as is more common in Alzheimer disease. Original magnification $\times 30$. **d** | Tendency for perivascular tau deposition and neurofibrillary tangle formation in the frontal cortex. Original magnification $\times 60$. Permission obtained from the American Medical Association © Shively, S. *et al. Arch. Neurol.* **69**, 1245–1251 (2012).

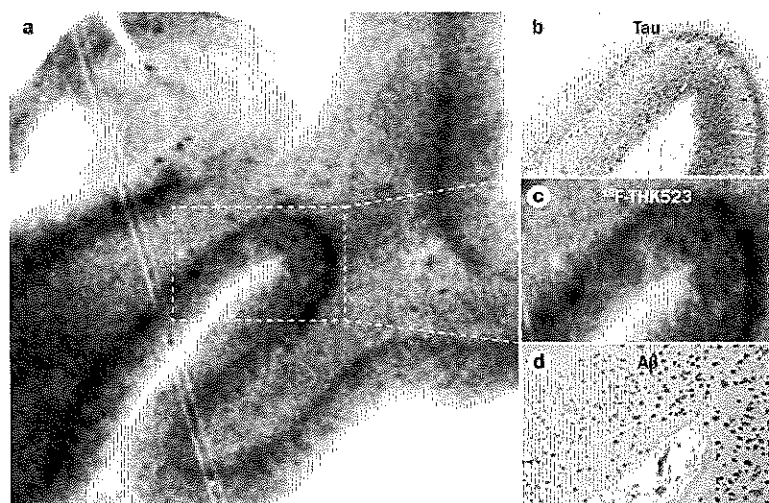


Figure 4.

¹⁸F-THK523 as a tauopathy marker. Autoradiography and microscopy analysis of hippocampal serial sections taken at autopsy from a 90-year-old patient with Alzheimer disease indicates that ¹⁸F-THK523 binds specifically to tau tangles, with no detectable binding to Aβ plaques. **a** | Low-magnification ¹⁸F-THK523 autoradiogram. **b–d** | Higher-magnification microscopy and autoradiogram images of three serial sections immunostained with antibodies to tau (**b**), which label neurofibrillary tangles; with ¹⁸F-THK523 (**c**); or with antibodies to Aβ (**d**), which label Aβ plaques. ¹⁸F-THK523 labelling seems to colocalize with tau immunostaining of neurofibrillary tangles, but not with plaques. Abbreviation: Aβ, amyloid-β. Permission obtained from Oxford University Press © Fodero-Tavoletti, M. T. *et al. Brain* **134**, 1089–1100 (2011).

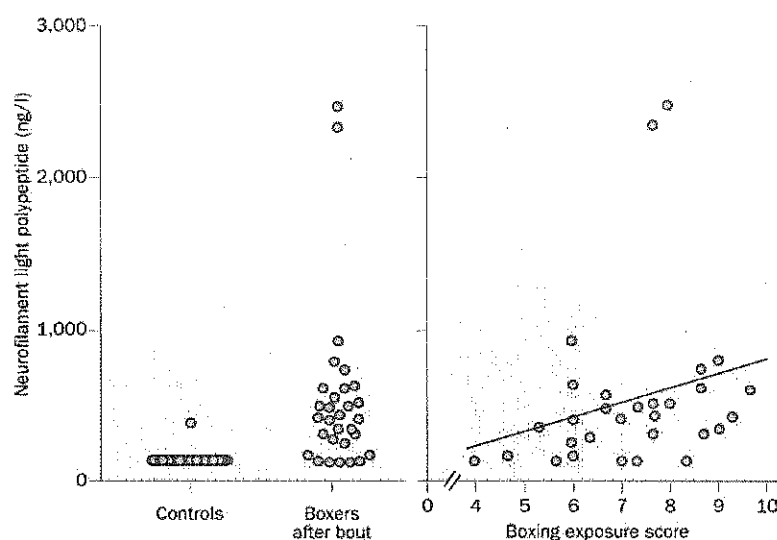


Figure 5.

CSF neurofilament light polypeptide in Olympic boxers after a bout. **a** | Individual CSF levels of neurofilament light polypeptide—a biomarker of axonal damage—in boxers after a bout and in age-matched controls. 25 of 30 (83%) of boxers had high CSF neurofilament light polypeptide levels, whereas 24 of 25 (96%) of controls had normal levels (below 125 ng/l), Wilcoxon signed rank test: $P < 0.001$. **b** | Correlation between boxing exposure and CSF neurofilament light polypeptide levels. A lower score indicates fewer and easier fights, whereas higher scores indicate more, tougher fights with more head blows. Spearman's $r = 0.40$; $P = 0.03$. Abbreviation: CSF, cerebrospinal fluid. Figure is reproduced from Neselius, S. *et al. PLoS ONE* 7, e33606 (2012), which is published under an open-access license by the Public Library of Science.